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Quantifying changes in ecological function of headwater catchments following large-scale surface mining in southern West Virginia

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Quantifying Changes in Ecological Function of Headwater Catchments Following Large-scale Surface Mining in Southern West Virginia

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Key words: decomposition, dissolved organic carbon, mitigation, ecological units, coal mining, amphibian colonization, macroinvertebrate colonization, surface mine reclamation, wetland creation, elevated conductivity and total dissolved solids

ABSTRACT

Quantifying Changes in Ecological Function of Headwater Catchments Following Large-scale Surface Mining in Southern West Virginia

Gretchen Anne Gingerich

West Virginia is one of the leading producers of coal in the United States. Large scale surface mining (mountaintop removal mining, MTR) has become commonplace in WV as a technique for accessing thin layers of coal that may be difficult or impossible to access through traditional underground mining techniques. MTR disturbs large areas of land and often results in a complete rearrangement of headwater catchments. Although restoring original pre-mining conditions is ideal, it is usually unrealistic in this region. Consequently, the extent to which post-reclamation watersheds function relative to the pre-mining conditions is unclear. The objectives of this study were to 1) quantify the functional value of reference headwater streams and post-reclamation aquatic features, 2) determine whether ecological functions are adequately replaced after mining and reclamation, and 3) develop recommendations for mining reclamation and direction for future studies.

Typically during mining and reclamation, steep, forested, ephemeral and intermittent stream channels that existed pre-mining are replaced with unforested, gently rolling terrain. Intermittent and perennial aquatic channels develop along the mine perimeter. These channels are constructed as sediment control complexes during the mining and mine-reclamation process. These aquatic features remain on the landscape post-reclamation and were the focus of this study's functional evaluation. Elevated metal concentrations were observed during some seasons at some mined locations. Overall, however, the reclamation process appears to do a good job of controlling metal contamination in water runoff. Perimeter channels, however, produced significantly higher levels of alkalinity, calcium, iron, magnesium, sulfate, specific conductivity, and total dissolved solids (TDS). Previous studies have shown these parameters to have significant downstream impacts on aquatic communities. These parameters are difficult to treat on-site and may be best managed at a watershed scale through the protection of undisturbed headwater catchments.

Over time, the reclaimed perimeter channels become vegetated with obligate wetland species, creating a considerable difference between mined and reference channels with regard to vegetation assemblages, canopy cover, and aquatic habitat quality. Species richness of macroinvertebrates and amphibians remains comparable between mined and reference channels. However, there is a distinct shift from sensitive, lotic taxa to tolerant, lentic taxa.

Reclaimed perimeter channel sites have a reduced ability to breakdown organic matter (OM), most likely as a result of reduced mechanical abrasion and reduced microbial activity due to elevated conductivity. Nevertheless, mined channels have significantly higher OM retention than reference channels. Consequently, perimeter channels show significantly higher overall processing power than reference channels. In other words, OM that enters a perimeter channel is retained and processed locally at a higher rate, whereas a greater proportion of OM entering a reference channel is transported downstream before being processed. As a result of higher OM retention and processing power, perimeter channels exported significantly more winter dissolved organic carbon (DOC) than reference stream sites.

This study represents a snapshot of relatively young (3 to 20 year old) reclaimed sites. Changes in vegetation, from open grassland to closed canopy forests, are expected as plant succession occurs over time. In addition, leaching of soluble salts may moderate many of the noted parameters such as alkalinity and sulfate that constitute high TDS values. Still, little is known about the rates and controlling factors of temporal changes.

Overall, a combination of on-site reclamation, off-site mitigation for lost structural components, and compensation at a watershed scale may be the best solution for shortcomings in current permitting and reclamation processes. Protecting native stream channels within mining-impacted watersheds would serve both as a source of dilute fresh water to compensate for alkaline drainage parameters and as a safeguard against the regional extinction of sensitive taxa.

Key words: decomposition, dissolved organic carbon, mitigation, ecological units, coal mining, amphibian colonization, macroinvertebrate colonization, surface mine reclamation, wetland creation, elevated conductivity and total dissolved solids

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Chapter 1

Quantifying Changes in Ecological Function of Headwater Catchments Following Large-scale Surface Mining in Southern West Virginia

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1.0 Introduction

1.1 Surface Mining and Reclamation

West Virginia is a leading producer of coal in the United States. Effectively managing the development of large-scale surface mines in southern West Virginia may be one of the most pressing environmental issues in the United States at this time. Large scale surface mining, commonly known as mountaintop removal mining (MTR), generates substantial volumes of excess spoil which are placed in external fills, permanently altering headwater streams. Individual fills may range from less than 25 hectares to hundreds of hectares and extend for thousands of meters in the watershed. The West Virginia Department of Environmental Protection (WVDEP) Code of State Rules stipulates "…no significant adverse impact to the chemical, physical, hydrologic, or biological components of aquatic ecosystems shall be allowed" (WVDEP 2007). Despite reclamation efforts, there remains considerable uncertainty over whether aquatic features on reclaimed mines are fulfilling Clean Water Act standards or are effective in recovering lost headwater functions.

1.2 Downstream Effects of Mining-related Disturbance

Several studies have been conducted on the effects of large-scale disturbances including deforestation, urbanization, and mining. There is some agreement about the general effects of these on local hydrology. Generally expected is an increase in runoff, stream flashiness, nutrients, relative abundance of exotic species, erosion potential, conductivity, sedimentation, metals, discharge, totals suspended and dissolved solids, and temperature (Likens et al. 1970, Dick et al. 1983, Martin et al. 1984, Bonta 2000, Tiwary

2001, Wiley et al. 2001, Bonta & Dick 2003, Wiley & Brogan 2003, USEPA 2005, Hartman et al. 2005, Maloney 2005, Meyer et al. 2005). Additionally, a decrease in organic matter export, streambed stability, macroinvertebrate richness and diversity, and pH may also be expected (Likens et al. 1970, Dick et al. 1983, Blevins 1991, Hartman et al. 2005, Maloney 2005, Meyer et al. 2005, Pond et al. 2008).

One of the largest issues resulting from disturbance caused by surface mining is the increase in total dissolved solids (TDS), salinity, and specific conductivity (all of which are highly correlated) in the resulting surface and groundwater. Efforts to remove or reduce TDS focus mainly on creating settlement ponds. Often the size of the sediment pond is significant enough to allow intentional or un-intentional hydrophytic vegetation growth. High TDS may negatively influence the biota of an aquatic feature. Spieles and Mitsch (2000) found that mean diel dissolved oxygen and specific conductivity were the best environmental predictors of invertebrate community metrics. Elevated conductivity, particularly, has been shown to interfere with the osmoregulation of macroinvertebrates (Wichard 1973, McCulloch 1993). Specific conductivity explained 16.4% of the differences in community structure (Spieles & Mitsch 2000). Ephemeroptera, specifically, have been shown to display the greatest response to increases in specific conductivity in waters affected by surface mining within the Appalachian region (Hartman et al. 2005, Pond et al. 2008).

Threat from acid mine drainage (AMD) exists when the pyritic minerals in coal and overburden are exposed to water and oxygen. The water leaving these sites is often highly acidic with high sulfate and monomeric aluminum concentrations (Baker et al.

1996, Merovich et al. 2007). When this water enters native catchments, aquatic ecosystems experience biological degradations such as altered periphyton community (Meegan & Perry 1996), reduced macroinvertebrate species richness, density, and biomass (Krueger & Waters 1983), and altered fish community assemblage (Krueger & Waters 1983, Baker et al. 1996, McClurg et al. 2007).

Macroinvertebrates are ubiquitous in natural settings and display diverse responses to environmental stresses. In part because of the community response, the analysis of benthic macroinvertebrates is generally accepted as a good measure of stream health (Purcell et al. 2002, Merovich & Petty 2007). Mining related impacts have been shown to result in a degradation of macroinvertebrate community structure and shifts in community composition primarily through an overall reduction in sensitive macroinvertebrate taxa (Garcia-Criado et al. 1999, Kennedy et al. 2003, Pond et al. 2008, Merriam 2009).

MTR and valley fill construction have also been shown to result in general degradation of stream fish communities. Stauffer and Ferreri (2002) found reduced fish species and reduced benthic fish species after mining-related impacts. Fulk et al. (2003) also found a reduction in overall Index of Biotic Integrity (IBI) scores in sites associated with mining. This was largely due to a reduction in minnow species and benthic insectivores.

Despite documented negative impacts of mining to downstream ecosystems and water quality, debate ensues over whether these impacts could, or should, be considered sources of impairment.

1.3 Cumulative Impacts of Headwater Functional Losses

Broad concern also exists over the effects of cumulative loss of headwater ecosystem functions from multiple mined watersheds. Over time, the cumulative effects of loss of headwater functions could cause unacceptable impacts to larger waterbodies downstream (Zedler 2003, USEPA 2005).

Previous literature has emphasized the connectivity of upstream functional and ecological processes to downstream ecosystem function and value (Vannote et al. 1980, Gomi et al. 2002, Lowe et al. 2006, Meyer et al. 2007, Wipfli et al. 2007). Generally, the ecological functions of individual headwater streams include the transport of invertebrates and organic matter and providing important trophic linkages (Wipfli 2005). Allochthonous headwater streams form an essential linkage in energy flow from upland forested watersheds to broad, turbid rivers downstream (Vannote et al. 1980). Fisher and Likens (1973) estimated that 66% of the annual energy budget of a second order stream was exported downstream. Native channels act as a source of coarse and fine particulate organic matter (CPOM/FPOM) for downstream trophic webs (Cummins & Klug 1979, Vannote et al. 1980, Cummins et al. 1989, Wallace et al. 1997). The losses of these energy sources may negatively affect overall productivity in downstream habitats.

Headwaters also act as refugia and source populations for downstream assemblages (Lowe & Bolger 2002, Meyer at al. 2007). Disturbed sites often benefit from the presence of these undisturbed sites as a source of re-colonization (Brown & Kodricbrown 1977). However, re-colonization may be affected by both the extent of the watershed affected and to the life histories of at-risk species to determine regional habitat needs (Lowe et al. 2006). Ensuring the connectivity of first-order streams may be essential for ensuring the survival of some species, such as *Gyrinophilus porphyriticus* (spring salamander) (Lowe & Bolger 2002, Lowe et al. 2006).

Additionally, the watershed-scale cumulative effects of mining may be confounded by the presence of other stressors within the watershed. Merriam (2009) found additive negative effects of mining and residential development on water chemistry and habitat degradation. Some studies show no cumulative effects of mining within watersheds on factors such as macroinvertebrate community structure (Fulk et al. 2003, Pond et al. 2008). However, studies have documented significant relationships between the density of mining within a watershed and the degradation of physical, chemical and biological conditions in streams (Maret & MacCoy 2002, Maret et al. 2003, Bruns et al. 2005). Although there is potential for such effects, the reality of this concern is currently largely undocumented.

1.4 Current Management Practices

Management techniques and best management practices (BMPs) exist to manage mining related impacts such as AMD production, sediment control, loss of vegetation, and increased concentration of water chemistry parameters. AMD is largely avoided both by restriction in the permitted locations of coal extraction and by material handling BMPs (Johnson & Hallberg 2005). Historically produced AMD is commonly mitigated for using limestone treatments (Weatherley 1988, McClurg et al. 2007).

On-site reclamation is often used to prevent, or lessen, potential downstream impacts. The purpose of reclamation is to improve the quality of the land by restoring pre-disturbance function (Bradshaw 1984). Additionally, reclamation seeks to reduce threats to downstream ecosystems such as impairment in water chemistry, channel sedimentation, impairment of riparian vegetation and aquatic habitat, and alteration to hydrology (primarily to peak flows) (Halverson & Sidel 1997). Surface mine reclamation became mandated in 1977 with the passage of the Surface Mining Control and Reclamation Act (SMRCA). Since that time, return of the site slopes to approximate the original contours and re-vegetation of the site to "a diverse, effective, and permanent vegetative cover of the same seasonal variety and native to the area and capable of self-regeneration and plant succession" within 5 years of mining has been required for bond release. Additionally, in terms of function, SMRCA requires "those actions taken to restore mined land to a post-mining land use approved by the Division of Mined Land Reclamation." Reclamation, however, does not imply the return of an ecosystem to an original state but does imply return to a functional state (Bradshaw 1996).

Erosion Control

According to Nicolau (2003), reproduction of the original topography of an area during reclamation is considered the geomorphic ideal; however, it is not always appropriate in steeper areas. Most surface mining in West Virginia occurs in steep, forested, headwater areas. Mining and filling these areas removes the forested cover and may alter the gradient. In the case of mountain top removal mining, steep, forested headwater streams are replaced by herbaceous vegetation, unconsolidated fill, and erosion control features. Typical erosion control consists of boulder-filled groin ditches and perimeter sediment ponds. These sediment ponds may be isolated or connected in series. They may or may not have downstream outflows. Often the size of the sediment pond is significant enough to allow intentional or un-intentional hydrophytic vegetation growth. Atkinson and Cairns (1994) found that of 14 “accidentally-formed” wetlands in a coal mined area in the Appalachians, all contained at least 26 obligate wetland plant species.

Although some studies have recommended both revegetation and removal of sediment ditches to reduce chemical load rates after reclamation (Bonta & Dick 2003), other authors encourage wetland construction in mined areas (Wieder & Lang 1984, Cole & Lefebvre 1991, Atkinson & Cairns 1994, Horstman et al. 1998). In several studies, constructed wetlands, particularly emergent wetlands, were found to support substantial waterfowl, anuran, and macroinvertebrate diversity (Horstman et al. 1998, Balcombe et al. 2005a,b). Wetlands were also found to be sinks for nitrogen, phosphorus, and carbon depending on their location in the watershed, residence time of water, and the volume of discharge (Raisin et al. 1997, Whiting & Chanton 2001). Results suggest rapid recovery of a watershed from disturbance. Constructed wetlands, in particular, demonstrate the botanical and biogeochemical characteristics of natural wetlands within a short time (less than 10 years), perhaps even within three to four years (Cole & Lefebvre 1991, Karamat et al. 1998, Maloney 2005). However, many would argue that a substantially longer period is more likely (Roberts 1993, Malakoff 1998, Zedler 2004).

<u>Water Quality</u>

Increasingly, retention ponds, or wetlands, are constructed in hopes of reducing unwanted water quality parameters such as dissolved metals (Sobolewski 1999).

Wetlands have been shown to be effective at removing aluminum, arsenic, cadmium, cobalt, copper, cyanide, iron, lead, manganese, nickel, uranium, and zinc (Sobolewski 1999). Sobolewski (1999) found that shallow depth and large inputs of organic matter were key characteristics of wetlands effective at removing metals from the water column.

Aquatic macrophytes play an essential role in removing metals from the water column by creating an environment suitable for metal removal (Brix 1994); but macrophyte removal usually accounts for only a minor proportion of total mass removed (Sobolewski 1999). Of macrophytes that are known to volunteer-establish on reclaimed and abandoned surface mines, Jones et al. (1993) found the average number of species per site was 18 with a maximum of 34 and a minimum of 7. Of those species, the rank of importance by cover and frequency placed *Typha sp., Scirpus cyperinus, Juncus acuminatus,* and *Juncus effusus* in the top four. Fortunately, metal removal by wetland ecosystems is not restricted to a particular plant species or climate (Prasad & Freitas 1999, Sobolewski 1999). Sobolewski (1999) did find that the pH of the drainage itself may influence the effectiveness of the wetland at removing metals, with acidic drainage impeding metal removal.

Microbial activity within the wetland plays an important role in metal removal (Sobolewski 1999). Microbial activity, combined with oxygen released from plant roots, creates an environment containing both aerobic and anaerobic conditions that allows for oxidation and reduction reactions simultaneously within the same location (Brix 1994). This allows metals to be removed from solution and retained in the sediments in stable, biologically unavailable forms (Sobolewski 1999).

Restoring Vegetation

Obstacles to establishing vegetation on mined sites include low pH (Haufler et al. 1978, Stocum 1980), soil compaction (Daniels & Amos 1981), elevated surface temperatures (Deely & Borden 1973, Stocum 1980, Bell & Unger 1981), and lack of nutrients such as nitrogen and phosphorus (Haufler et al. 1978, Andrews 1992). Additionally, colonization by native plants may be hindered by distance to intact forests and sources of seed, soil compaction by machinery (Davison et al. 1984), and competition by intentionally planted non-native plants (Connell & Slatyer 1977). After 35 years post mining, Holl (2002) found reclaimed mine sites differed substantially from reference sites in terms of vegetation.

Depending on the species, initial vegetation establishment may either encourage, not effect, or inhibit additional species establishment (Connell & Slatyer 1977). Reclaimed sites are often seeded with rapidly growing non-native species, such as the grass *Festuca arundinacea*, for erosion control purposes. These species may hinder succession by native species and overall ecosystem recovery (Brenner et al. 1984, Burger & Torbet 1990, Hughes 1992, Chambers et al. 1994, Holl & Cairns 1994, Skousen et al. 1994, Torbet et al. 2000, Allen et al. 2001, Skousen et al. 2006).

Reclaimed mine sites are often planted with *Pinus strobus* (eastern white pine), a pine often native to mined catchments. Although *P. strobus* is effective at increasing tree basal area on site, it may hinder establishment of herbaceous vegetation as the canopy becomes more closed (Ashby 1964, Schuster & Hutnik 1987, Holl 2002). Additionally, pine plantations have lower wildlife value than hardwood stands (Leedy 1981).

However, establishment of tree species in general is advised as some vegetative species may be unable to colonize without presence of a canopy (Holl & Cairns 1994).

Holl and Cairns (1994) found the number of species increased with age since reclamation. Other authors found a large number of native species had colonized reclaimed surface mines after 10-15 years (Thompson et al. 1984, Skousen et al. 1994, Thompson et al. 1996, Rodrigue 2001). Often reclaimed sites are dominated by generalist species such as *Acer rubrum* (Holl 2002) and species dependent on insect activity or gravity for seed dispersal are not present (Holl 2002, McLachlan & Bazely 2001). Holl (2002) estimates a time period of over 35 years before reclaimed sites host a vegetative community fully comparable to reference sites.

Suggestions for the improvement of vegetation establishment include keeping reclaimed mine sites within 50 meters from intact forest to provide a seed source (Holl 2002), additional research on naturalized ground covers such as wildflowers that would not impede vegetation succession (Holl 2002), planting a variety of tree species for a diverse canopy (Holl 2002), improvement of within-site environmental conditions (Holl & Cairns 1994), and movement of soil from imminently mined locations to reclaimed sites (Garrison 1992).

Parameters that cannot be reclaimed on-site are often mitigated for off-site usually through habitat enhancement structures and channel reconfiguration techniques that seek to improve water quality, in stream habitat, and bank stabilization (Bernhardt et al. 2007).

1.5 Reclamation Challenges

Despite BMPs, challenges in mining reclamation still exist. The production of AMD and the export of sediments are largely avoided but high TDS and conductivity concentrations remain and are exported downstream (Pond et al. 2008, Minter 2009, Merriam 2009). Additionally, the ecological functions of reclaimed mine sites are largely unmeasured and the effectiveness of off-site mitigation efforts to recapture discrepancies in function may be questioned.

1.6 Remaining Questions

Numerous important questions remain. To what extent do reclaimed headwater watersheds function in comparison to native headwater watersheds? Which ecosystem structures and functions may be completely lost despite reclamation? Which functions are significantly reduced? Which functions are comparable? Which functions are increased? Which functions may improve with time? Do techniques exist to improve functional reclamation? Which remaining functional deficits need to be addressed through off-site mitigation?

2.0 Objectives

The overriding objective of this research project was to quantify and contrast ecological function of reference aquatic features to post-reclamation aquatic features that develop on reclaimed surface mines in southern West Virginia. The long-range goal is to maximize the functional value of on-site aquatic features and the recovery of lost headwater functions throughout the Appalachian region. In this study, we addressed the following specific objectives:

<u>Objectives:</u>

1. quantify the functional value of reference headwater streams and of post-reclamation aquatic features;
2. determine whether ecological functions are adequately replaced after mining and reclamation; and
3. develop recommendations for mining reclamation and direction for future studies.

3.0 Methods

3.1 Study Area

Perimeter channel sites consisted of five boulder-filled, sediment control structures on the perimeter of reclaimed surface mine lands in the coal-rich region southwest of Charleston, WV (Fig. 1). Within the region, typical post-reclamation surface mine structures are composed of a re-contoured and re-vegetated “on-bench” site located adjacent to, and at greater elevation than, adjacent valley fills associated with the site. The chosen sites varied in drainage area and age since reclamation but all were designed so that any overland flow from reclaimed mine lands drained towards the perimeter of the site and into the perimeter channels. All sites drained towards an “on-bench” outflow notch in the berm that surrounded the reclaimed surface mine perimeter. Figure 2 illustrates a typical reclaimed mine perimeter channel series in comparison to a typical undisturbed headwater system. Study reaches within the sites began one retention cell above the channel series off-site outflow notch and continued upstream a length of 10 times the mean channel width. If the outflow notch was located at the confluence of two perimeter channels, the wetter channel series was chosen.

These features were constructed primarily for the purpose of slowing and retaining water to allow the settling-out of suspended sediments and some ions from the water column before discharge of water off-site. Information on estimated age since reclamation was included in the study, however, no information on the site's history of maintenance dredging and re-contouring since reclamation was obtained. Therefore, overall age since reclamation may not represent a continuous successional trajectory. Additionally, perimeter channel sites were not intentionally designed to encourage ecological function. Any vegetation growth or colonization by macroinvertebrates or amphibians was not an intended use of the structure.

Reference sites consisted of five intermittent streams within the region that were of the best sites available within a reasonable proximity to perimeter channel sites. These were relatively unaffected by disturbance and accessible by road. Sites were selected using winter and spring 2008 water chemistry measurements as well as topographic maps and general knowledge of the area. Reach length was measured at 10 times the mean stream width with a minimum length of 50 m. A list of site names, watersheds, and drainage areas are given in Table 1.

3.2 Physical Habitat

Habitat Quality

Habitat analysis was performed using classification systems such as Virginia Unified Stream Method (USACOE 2007), West Virginia Functional Channel Unit Assessment (USACOE & VADEQ 2007), Wildland Hydrology's Bank Erosion Hazard Index (Rosgen 2001), Environmental Protection Agency Rapid Bioassessment Protocol (Barbour et al. 1999), and Ohio Rapid Assessment Method (Mack 2001). These systems use simple measurements and visual assessments to assign sites numerical scores to each site. Categorical habitat qualifiers (e.g. excellent, good, poor) were derived from calculated scores.

Vegetation

Vegetation was sampled according to protocols adapted from Batzer et al. (2004), Balcombe (2005, b), and Rentch et al. (2008). Thirty meter long transects were run across the site perpendicular to the length of the stream or perimeter channel. A pin flag was used every 2 m along the transect to record any intercepting vegetation. Additionally, fifteen 1x1 m plots were placed along each transect. Within each plot, percent cover was estimated using 14 categories (bare ground, cattail, exposed substrate, fern, forb, grass, moss, open water, rush, sedge, shrub, submerged vegetation, tree, and vine). All course woody debris within the plot was tallied and recorded. Canopy cover was estimated using a spherical densitometer. Diameter at breast height (dbh) and species was recorded for any trees > 2.5 cm dbh and within 10 m of the transect. Any additional species not captured in survey measurements were noted.

3.3 Water Chemistry, Temperature, and Discharge

Water Chemistry

Sites were visited seasonally from February 2008 to May 2009. A list of sample dates is given in Table 2. Instantaneous water chemistry measurements were taken at each visit with a Yellow Springs Instrument (YSI) 650 multi-parameter probe equipped with a 600XL sonde. The YSI was calibrated before every site visit. The probe measured temperature, pH, conductivity, and dissolved oxygen.

Seasonal water samples were taken to analyze dissolved water chemistry including metals and nutrients following protocols by Merovich et al. (2007). Grab samples were taken by completely filling a sampling bottle under the water's surface. Filtered samples were taken using a vacuum pump and mixed ester cellulose membrane 0.45 µm filter. Nitric acid (HNO_3) was added to keep all solutes in solution. Analysis was performed by the National Research Center for Coal and Energy at West Virginia University, Morgantown, WV. Alkalinity was measured in $CaCO_3$ equivalents and presented as mg/L. If samples were measured below the method detection limit (MDL), one half of the MDL was used in analysis. Samples measuring below MDL are presented as "MDL" in figures. Mean seasonal data was calculated using three seasons only (spring, summer, and winter) because of the absence of water during autumn sampling.

Specific limits for parameters were taken from West Virginia guidelines for warm water fisheries and comply with EPA regulations. For limits dependent on hardness, a hardness of 0-50 mg/L was used to determine limits. Total dissolved solids (TDS) was calculated by summing seasonal measurements for dissolved ions.

Water Temperature

Hourly temperature readings were taken from June 2008 until June 2009 using HOBO U22 Water Temp Pro v2 loggers (Onset Computer Corporation, Pocasset, MA). No temperature readings were recovered from P_WO before the temperature logger was buried by bulldozer activity. Loggers for R_WO and R_MW ceased logging prior to June 2009 and resulted in incomplete data. Data from periods when loggers were inundated were used for analysis.

Discharge

Discharge was calculated from width, depth, and flow measurements taken at each site visit. Flow was measured using a Flow Mate 2000 flow meter. Measurements were summarized by season for analysis.

3.4 Biotic Components

Amphibians

Amphibians were sampled seasonally in early and late spring and early and late summer (March, May, June, and July) of 2008. This series of sampling sought to maximize the diversity captured for each site as well as sample during all potential breeding seasons for probable amphibian species. Adult assemblages were estimated using Visual Encounter Surveys (VES) performed in accordance with protocols set by Crump and Scott (1994). Larval Surveys were adapted from methods by Shaffer et al. (1994). The surveys were comprised of consistent meter-long sweeps with a D-frame net in open-water and consistent meter by half-meter area searches in stream channels. Each search was done at 10 random locations over the length of the site.

Macroinvertebrates

Macroinvertebrates were sampled in spring and fall 2008. Within lotic systems, macroinvertebrates were sampled using protocols established by Merovich and Petty (2007), which are slight modifications of procedures described by the West Virginia Department of Environmental Protection's Watershed Assessment Program and the EPA's Rapid Bioassessment Protocols for wadeable streams (WVDEP 1996, Barbour et al. 1999). Within each reach, four representative riffles were sampled using a 250 µm D-frame kicknet. Within lentic systems, a D-frame net was used to take jab samples at 10 random locations along the reach. Lentic samples were taken according to protocols set by King and Richardson (2002) and Balcombe et al. (2005a). Samples were preserved with 70% ethanol and identified to the lowest possible taxonomic level. Fall 2008 samples were obtained in perimeter sites only due to low water in reference sites. Consequently, we analyzed data from spring samples only.

Stream condition was derived from macroinvertebrate samples using a modified West Virginia Stream Condition Index (WVSCI) score developed by Merriam (2009) from original Gerritsen et al. (2000) protocols. This index uses family-level community metrics to categorize stream condition as either poor, marginal, good, or excellent.

3.5 Organic Matter Processing

Organic Matter Retention

OM transport was measured using methods adapted by Minter (2009) from protocols established by Speaker et al. (1984), Webster et al. (1994), Raikow et al. (1995), and Lamberti (1996). Cumulative retention was measured using artificial sticks, consisting of painted dowel rods, and instantaneous retention was measured using artificial leaves, consisting of rectangles of blue construction paper. Fifty dowel rods were placed in a riffle at the upstream end of the reach. They remained on-site and the cumulative distance they traveled was measured at four intervals over 195 days (on the day of release and then after approximately 1 month, 2 months, and 6 months). Twenty artificial leaves were also placed in a riffle at the upstream end of each reach seasonally. They were allowed to travel downstream for 30 minutes then their distances traveled were recorded.

Retention rate was calculated for each site using the cumulative distances traveled by artificial sticks. The equation $T_d = T_o\ e^{-kd}$, where T_d is the percentage of released sticks remaining in transport at distance d, T_o is the original number of sticks released, and k is the instantaneous rate of retention, was used to calculate retention rate (Speaker et al. 1984, Raikow et al. 1995, Minter 2009).

<u>Dissolved Organic Carbon</u>

Seasonal water samples were taken from each site. Samples were filtered with a 0.45 µm filter and treated with nitric acid (HNO_3). Samples were analyzed using a Sievers 5310c laboratory TOC Analyzer to estimate total dissolved carbon and dissolved organic carbon concentrations.

<u>Organic Matter Decomposition</u>

Leaf litter packs were constructed from plastic mesh bags (10 mm mesh size). This allowed the bag to hold material in one location without restricting access to the material by invertebrates or breakdown of the material by water flow. Bags were filled with 10 g of *Quercus palustris* (pin oak) leaves collected after abscission and air-dried for approximately two weeks to a constant mass. Bags were grouped in sets of six and anchored in riffles throughout the reach length. An additional set of litter bags was taken to the site and returned to the lab to calculate for handling loss. Litter bags were randomly sampled after 45, 75, 90, 120, 195, and 325 days on site. Bags were returned to the lab on ice and rinsed in a 250 µm sieve. Macroinvertebrates present in litter bags were collected, preserved, and identified. Leaf litter was placed in brown paper bags and dried for approximately 48-72 hours to a constant mass. After drying, leaf litter was reduced to particulate size and subsampled. A subsample of 250 µg was placed into pans and incinerated to determine the ash free dry mass. Methods were adapted from protocols by Benfield (1996). Decomposition rates (-k) were determined from the linear regression of the plot of the number of days of exposure versus the log-transformed percent ash-free dry mass (% AFDM). AFDM was calculated using the following formula from Benfield (1996):

(Equation 1) $\%\ \text{Organic Matter} = (DM_{sample} - AM_{sample}) / DM_{sample} \times 100$

$AFDM = DM \times \%\ \text{Organic Matter}$

$\%\ AFDM\ \text{remaining} = 100 - ((\text{initial} - \text{final}) / \text{initial} \times 100)$

Where: DM = dry mass; AM = ash mass

The slope of the line after regression provided the rate of decomposition (k). Expected decomposition rates for *Quercus* range from k= (0.0014) to k= (0.021) (Beiser et al. 1991, D’Angelo & Webster 1992).

Processing Power

Processing power was defined as the ability of a site to retain and process organic matter locally. Processing power of each site was calculated by multiplying the instantaneous rate of decomposition by the instantaneous rate of retention.

3.6 Site Comparison

Because pre-mining reference sites were intermittent streams and post-reclamation perimeter channels sites resembled wetlands, measured parameters differed in terms of how directly comparable they were. Some parameters such as water chemistry and decomposition rate are directly comparable between site types (Fig. 3). However, some parameters such as gradient and vegetation community may not be directly comparable between site types.

3.7 Ecological Units

Ecological units (EUs) (Petty & Thorne 2005, Merovich & Petty 2007) were calculated for parameters selected as important metrics for both perimeter and reference site types. They were calculated by dividing the perimeter mean by the reference mean for each parameter. EUs with values greater than one represent ecosystem parameters with higher values on the perimeter channel sites than in reference catchments. EUs with values less than one represent ecosystem parameters with higher values in reference streams than in perimeter channels.

3.8 Statistical Analysis

Data were transformed using $\log_{10}$, arcsine, and square roots of measured values before analysis in order to approximate normality within the data. Correlation analysis was run on all parameters to quantify relationships between ecosystem parameters and both conductivity and time (with respect to perimeter channels only). T-tests were used to test for statistical differences in ecosystem parameters between site types (perimeter and reference). Repeated measures ANOVAs were used to test for seasonal variation in ecosystem parameters between perimeter and reference sites. ANCOVAs were used to test for interactive effects of site type and specific conductivity on ecosystem parameters.

Relation among macroinvertebrate and amphibian communities and environmental variables were examined using non-metric multidimensional scaling (NMDS). NMDS is a statistical ordination, developed by Clarke (1993). This non-parametric analysis involves iterative solutions that allow species composition data to be plotted in ordination space with reduced stress (McCune & Grace 2002). This analysis enables the determination of community similarities as well as the influence of environmental variables on community composition. Additionally, Spearman rank correlations were run between NMDS scores and community metrics (Merovich & Petty 2007, McClurg et al. 2007, Merriam 2009).

All statistical analyses were conducted using the program R Project for Statistical computing version 2.8.1 (R Development Core Team 2008) unless otherwise stated. NMDS analysis was run using the package vegan (Oksanen et al. 2008). All values were considered significant at an alpha level of 0.05 unless otherwise indicated.

4.0 Results

4.1 Habitat Quality

Habitat quality measures were consistently higher in reference channels than in perimeter channels regardless of the assessment protocol. EPA rapid bioassement protocol (RBP) scores averaged 78 for perimeter channels and 150 for reference sites (Table 3). Mean scores for the Virginia unified stream method (VA USM) were 4 for perimeter channels and 6 for reference sites. West Virginia functional channel unit (WV FCU) assessment scores averaged 3 for perimeter channels and 9 for reference sites. In the case of Wildland Hydrology's bank erosion hazard index (BEHI), the higher the score, the more prone a site is to erosion. BEHI scores averaged 23 for perimeter channels and 39 for reference sites. All habitat assessments were significantly higher in reference sites than in perimeter channels (Table 4).

The Ohio Rapid Assessment Method (ORAM) version 5.0 was designed to rank overall wetland quality and to categorize natural wetlands based on amount of disturbance (Mack 2001). The higher the score, the less disturbed the wetland. Perimeter channels averaged an ORAM score of 35 and reference sites averaged 61. ORAM scores were significantly higher in perimeter channels than in reference sites (Table 4).

4.2 Vegetation

Perimeter channel sites were dominated by forb (22%), grass (22%), cattail (21%), and open water (13%) (Table 5). Reference sites were dominated by bare ground (29%), forb (22%), trees (16%), and fern (11%). Mean percent canopy cover was 4% for perimeter channels and 91% for reference sites (Table 6). Perimeter channels had, on average 0.3 ± 0.6 trees and 0.1 ± 0.2 tree species per km^2 survey versus an average of 9.2 ± 5 trees and 2.5 ± 0.7 species per km^2 in reference sites. All vegetation measures were significantly different between site types except for percent open water and forb.

4.3 Water Chemistry

Parameters that Were Similar Between Site Types

pH

The majority of sites had mean seasonal pH measurements that fell within the range of 6.0-8.0 (Fig. 4). Mean summer pH was significantly higher in perimeter channels (7.39) than in reference channels (6.74) (Table 4). Greater pH variation occurred among reference sites than among perimeter channel sites. One reference site, R_WO, had consistently low pH ranging from a low of 4.35 in summer to a high of 5.82 in autumn. When excluding R_WO from the analyses, pH was similar between reference sites (7.15 excluding R_WO) and perimeter channel sites. No significant difference in mean pH was observed between reference and perimeter channel sites (Table 4).

Dissolved Oxygen

Dissolved oxygen (DO) levels below 5.0 mg/L stress aquatic life. Statistically significant trends were shown for DO in regards to season (Table 4). The seasonal mean for all sites was above the 5.0 mg/L level except R_ME which had a summer mean of 4.5 mg/L (Fig. 5). Perimeter channel sites averaged 8.7 mg/L and reference sites averaged 9.9 mg/L for the study duration. No significant difference in dissolved oxygen concentrations was observed between reference and perimeter channel sites (Table 4).

Dissolved oxygen did, however, show statistically significant trends with regards to seasonal concentration (Table 7).

Manganese

West Virginia water quality criteria in conformance with USEPA regulations limits manganese within warm-water fisheries (WWF) to a level of 1 mg/L. Most perimeter channels averaged above this limit and reference sites averaged below this level (Fig. 6). Reference site R_WO's spring, summer, and winter measurements were 1.87, 1.90, and 1.10 mg/L respectively. Mean seasonal measurement for perimeter channels was 0.3 mg/L. Mean seasonal measurement for reference sites was 0.1 mg/L excluding measurements from R_WO (0.4 mg/L for all reference sites). No significant difference in manganese concentrations was observed between reference and perimeter channel sites (Table 4). Seasonal water chemistry measurements for each site can be found in Appendix A.

Iron

The WWF limit for iron is 1.5 mg/L (Fig. 7). All sites were below this level. Mean seasonal measurements were 0.10 mg/L for perimeter channels and 0.06 mg/L for reference sites. Mean iron concentrations were not significantly different between site types (Table 4).

Zinc

The WWF level for zinc is 0.04 mg/L. Most site measurements were below these levels (Fig. 8). Autumn measures for P_BH (0.115 mg/L) and P_WO (0.141 mg/L) exceeded recommended levels. Seasonal measurements for perimeter channels averaged

0.020 mg/L and 0.042 mg/L for reference sites. No significant difference in zinc concentrations was observed between reference and perimeter channel sites (Table 4).

Selenium

WWF level for selenium is 0.005 mg/L. However, method detection limits (MDL) of the water sample analysis technique were 0.045 mg/L. All sites were at this detection level except for autumn measurements for P_ST (0.173 mg/L) and P_WO (0.148 mg/L) (Fig. 9). No significant difference in selenium concentrations was observed between reference and perimeter channel sites (Table 4).

Nitrite

The WWF limit for nitrite is 5 mg/L of nitrite. Most seasonal measurements for nitrite were below 0.1 mg/L (Fig. 10). Summer measurement for P_WO was 0.40 mg/L. Summer measurement for R_HC was 46.23 mg/L after disturbance. Mean seasonal measurements were 0.04 mg/L for perimeter channels and 0.02 mg/L for reference sites, excluding R_HC measurements (reference site mean was 3.10 mg/L including all sites). No significant difference in nitrite concentrations was observed between reference and perimeter channel sites (Table 4).

Nitrate

The WWF limit for nitrate is 90 mg/L. The majority of sites were below 20 mg/L (Fig. 11). Summer and autumn measurements for P_WO were 143.8 mg/L and 79.3 mg/L respectively. Mean seasonal measurements were 0.8 mg/L for reference sites and 0.6 mg/L for perimeter channels excluding P_WO measurements (perimeter site mean was 11.0 including all sites). No significant difference in nitrate concentrations was observed between reference and perimeter channel sites (Table 4).

Total Phosphorus

In 1986, the EPA recommended a phosphorus level of 0.1 mg/L for rivers not emptying into reservoirs. All sites were below this level except P_WO which measured 0.68 mg/L (winter) after disturbance (Fig. 12). Mean seasonal measurements were 0.07 mg/L (0.04 mg/L excluding P_WO) for perimeter channels and 0.05 mg/L for reference sites. No significant difference in total phosphorus concentrations was observed between reference and perimeter channel sites (Table 4).

Barium

All sites measured below 1.0 mg/L of barium for all seasons. Generally sites measured below 0.2 mg/L except R_HC which measured 0.887 mg/L in summer (Fig. 13). Mean seasonal measurements were 0.016 mg/L for perimeter channels and 0.093 mg/L for reference sites. No significant difference in barium concentrations was observed between reference and perimeter channel sites (Table 4).

Ammonia

The WWF limit for ammonia is 0.05 mg/L. All sites were below this level except for P_ST which measured 0.087 mg/L (autumn) and R_HC which measured 0.056 (summer) after disturbance (Fig. 14). Mean seasonal measurements were 0.007 mg/L for perimeter channels and 0.013 mg/L for reference sites. No significant difference in ammonia concentrations was observed between reference and perimeter channel sites (Table 4).

Cobalt

There are no recommended limits for cobalt. MDL for cobalt during analysis was 0.015 mg/L. Measurements for all sites were at the MDL except for P_BH (0.021 mg/L; autumn), P_ST (0.054 mg/L; autumn), P_WO (0.021 mg/L; summer), and P_SU (0.018 mg/L; summer) (Fig. 15). Mean seasonal measurements were 0.011 mg/L for perimeter channels and 0.009 mg/L for reference sites. No significant difference in cobalt concentrations was observed between reference and perimeter channel sites (Table 4).

Copper

The WWF limit for copper is 0.006 mg/L. MDL during analysis for copper was 0.015 mg/L. All sites were at MDL except P_BH which measured 0.027 mg/L (autumn) and P_ST which measured 0.061 mg/L (autumn) (Fig. 16). Mean seasonal measurements for analysis were 0.008 mg/L for perimeter channels and 0.008 mg/L for reference sites. No significant difference in copper concentrations was observed between reference and perimeter channel sites (Table 4).

Cadmium

The WWF limit for cadmium is 0.007 mg/L. However, MDL for cadmium was 0.014 mg/L. All sites measured at the MDL except P_BH which measured 0.023 mg/L (autumn) and P_ST which measured 0.057 mg/L (autumn) (Fig. 17). Mean seasonal measurements for analysis were 0.007 mg/L for perimeter channels and 0.008 mg/L for reference sites. No significant difference in cadmium concentrations was observed between reference and perimeter channel sites (Table 4).

Parameters that Differed Between Site Types

Acidity

Perimeter channels averaged 0 mg/L acidity for all seasons (Fig. 18). Reference sites averaged 13 mg/L over the duration of the study. Mean acidity was significantly lower in perimeter channels than in reference sites (Table 4).

Alkalinity

Waters with alkalinity measures above 20 mg/L are considered to have good buffering capacity. Alkalinity averaged 138 mg/L for perimeter channels and 5 mg/L for reference sites (Fig. 19). Mean alkalinity concentrations were significantly higher in perimeter channels than reference sites (Table 4).

Calcium and Magnesium

Mean seasonal measurements of calcium were significantly higher in perimeter channels (163 mg/L) than in reference sites (21 mg/L) (Fig. 20, Table 4). Streams with magnesium sources usually have levels of 5-50 mg/L. Mean seasonal measurements of magnesium were also significantly higher in perimeter channels (154 mg/L) than in reference sites (9 mg/L) (Fig. 21, Table 4).

Sulfate

Perimeter channels measured above 250 mg/L and reference streams measured below this level (Fig. 22). Mean seasonal measurements for perimeter channels were 1008 mg/L. Mean seasonal measurements for reference sites were 32 mg/L. Mean sulfate levels were significantly different between site types. Mean sulfate concentrations were significantly higher in perimeter channels than reference sites (Table 4).

Chromium

The WWF limit for chromium is 0.01 mg/L. However, MDL for chromium was 0.012 mg/L. All sites measured below 0.03 mg/L except P_ST which measured 0.066 mg/L (autumn) (Fig. 23). The mean seasonal measurements were 0.006 mg/L for perimeter channels and 0.009 mg/L for reference sites. Chromium concentrations were significantly higher in perimeter channel sites than in reference sites (Table 4).

Specific Conductivity and TDS

The U. S. Environmental Protection Agency (EPA) recommends a specific conductivity level of 150- 500 µS/cm for aquatic health. Mean specific conductivity exceeded the recommended level of 500 µS/cm at all perimeter channels for all seasons during the study period (Fig. 24). The mean measured specific conductivity for perimeter channels was 2197 µS/cm. Reference sites averaged below 500 µS/cm with the exception of R_HC which measured 2362 (spring) and 2632 (summer) due to an upstream disturbance. Mean specific conductivity for reference sites, excluding R_HC, was 141 µS/cm (overall mean was 461 µS/cm for reference sites). Mean conductivity levels were significantly higher in perimeter channels than reference sites (Table 4). Specific conductivity was positively correlated with mean alkalinity, discharge, calcium, iron, magnesium, sulfate, summer pH, TDS, percent macroinvertebrate predator, and percent open water (Table 4). Conductivity was negatively correlated with percent Ephemeroptera, Plecoptera, and Trichoptera (EPT), and EPT richness (Table 4).

Mean TDS was also significantly higher in perimeter channels than in reference sites (Table 4). Mean TDS measures were 1501 mg/L in perimeter channels and 103 mg/L in reference sites. TDS was composed primarily of bicarbonate, calcium, magnesium, sulfate, chloride, and sodium (Fig. 25). The mean percent of TDS composed of sulfate in perimeter channels was 66% and 43% in reference sites. TDS also showed significant trends in relation to seasonal concentrations (Table 7).

<u>Parameters with Unusual Values</u>

Aluminum

The WWF limit for aluminum is 0.75 mg/L. Seasonal measurements for all sites were below these levels except for R_WO (Fig. 26). R_WO measured 3.0 mg/L (spring), 3.1 mg/L (summer), and 1.3 mg/L (winter). Mean seasonal measurements were 0.1 mg/L for perimeter channels and 0.5 mg/L for reference sites. No significant difference in aluminum concentrations was observed between reference and perimeter channel sites (Table 4).

Nickel

The WWF limit for nickel is 0.088 mg/L. All sites measured below this level for all seasons except P_WO which measured 0.132 mg/L (summer) and 0.144 mg/L (winter) (Fig. 27). Mean seasonal measurements for perimeter channels were 0.034 mg/L (0.016 mg/L excluding P_WO) and 0.024 mg/L for reference sites. No significant difference in nickel concentrations was observed between reference and perimeter channel sites (Table 4).

Chloride

The WWF limit for chloride is 250 mg/L. Most seasonal chloride measures were below this limit (Fig. 28). Seasonal measures for P_BH were 43.1 mg/L (spring), 90.6

(summer), 111.2 mg/L (autumn), and 24.3 mg/L (winter). Seasonal measures for R_HC after disturbance were 1070.7 mg/L (summer) and 102.4 mg/L (winter). Mean seasonal measurements for perimeter channels were 13.8 mg/L (4.5 mg/L excluding P_BH) and 11.4 mg/L (1.17 mg/L excluding R_HC) for reference sites. No significant difference in chloride concentrations was observed between reference and perimeter channel sites (Table 4).

Sodium

All sites measured below 20 mg/L except P_BH, P_ST, and R_HC (Fig. 29). P_BH measured 40.3 mg/L (spring), 44.3 mg/L (summer), 61.4 mg/L (autumn), and 28.4 mg/L (winter). P_ST measured 20.9 mg/L (spring). R_HC measured 293.2 mg/L (summer) and 44.0 mg/L (winter) after disturbance. Mean seasonal measurements were 7.8 mg/L for perimeter channels, excluding P_BH (13.2 mg/L including all sites), and 1.6 mg/L for reference sites, excluding R_HC (23.7 mg/L including all sites). No significant difference in sodium concentrations was observed between reference and perimeter channel sites (Table 4).

4.4 Temperature

Continuous temperature loggers providing a year's worth of data showed perimeter channels to have a similar temperature range to reference sites (10.44 ± 0.56 °C vs 9.59 ± 1.67 °C respectively) (Table 4 & Table 8). There was no overall trend in temperature between site types (Fig. 30, 31 & 32). Statistically significant trends were shown for temperature in regards to season (Table 7). Seasonal temperature measurements are summarized in Appendix B.

4.5 Discharge

Statistically significant trends were shown for discharge in regards to season (Table 7). Seasonal discharge averaged higher in perimeter channels than in reference sites (0.00897 m^3/s vs. 0.00233 m^3/s respectively) (Fig. 33). Mean spring discharge was significantly higher in perimeter channel sites than reference sites (Table 4). Flows were highest for all sites in winter. All reference streams except R_WO had little to no flow from late summer through early autumn. Perimeter channel sites showed more variation between their seasonal averages than reference sites. Mean seasonal standard deviation was ± 0.0089 m^3/s for perimeter channels and ± 0.0021 m^3/s for reference sites.

4.6 Amphibians

Perimeter channels contained, on average, more larval amphibians than reference sites (avg 19 vs 8 individuals) and supported about the same number of species (avg 2 species) (Table 9). Mean larval biomass also averaged higher in perimeter channels (1.33 g/m^2) than reference sites (0.05 $g/100m^2$) (Table 10). Perimeter channels contained, on average, less adult amphibians than reference sites (avg 5 vs 28 individuals) but supported a similar number of adult species (avg 2 vs 3 species). When the effects of conductivity were removed, larval amphibian richness, total number of larval amphibians, and the percent of lotic-utilizing amphibians was statistically different between site types with perimeter channels supporting more larva and reference sites supporting more lotic-utilizing species (Table 11). Overall density was not statistically different between perimeter channels and reference sites (Table 4, Table 12, Fig. 34). Amphibian survey data can be found in Appendices C-E.

Perimeter channels supported primarily terrestrial and aquatic frogs that use lentic systems. Reference sites supported primarily aquatic salamanders that use lotic systems (Table 9 & Table 13). The species of both site types were able to use forest habitats but perimeter channels supported a statistically significant higher percentage of grassland-utilizing species (Table 4). Additionally, perimeter channels supported a significantly lower percentage of lotic-utilizing species and a significantly higher percentage of lentic-utilizing species (Table 4). The percent of lentic species was positively correlated with conductivity and percent lotic species was negatively correlated with conductivity (Table 4).

NMDS ordination analysis revealed clustering by site types. Amphibian community structure was primarily influenced by vegetation and water chemistry parameters including percent open water, grass, cattail, bare ground, canopy cover, fern, and number of species per km^2 (Fig. 35). Increasing percent open water, grass and cattail indicated a perimeter- type amphibian community while increasing percent bare ground, canopy cover, fern, and species per km^2 indicated a reference- type community structure. Statistically significant water chemistry influences included mean specific conductivity, sulfate, magnesium, mean total dissolved solids, calcium, alkalinity, and iron (Fig. 36). Increasing measures of all these parameters indicated a perimeter- type community composition. Additionally, ANCOVA analysis revealed significant interactions between site type and site conductivity for amphibian species richness, larval richness, total number of amphibians, mean number of amphibians, number of larval amphibians, mean density, and percent forest-utilizing amphibians (Table 11, Fig. 38-42).

4.7 Macroinvertebrates

Perimeter channels and reference sites had similar macroinvertebrate family richness and biomass. Perimeter channels had an average of 8 families and reference sites had an average of 7 families (Table 14). Perimeter channels had an average biomass of 31.76 g/m^2 and reference sites had an average of 34.47 g/m^2. Perimeter channels had a higher percentage of tolerant species (70% vs 42%) and chironomids (58% vs 32%) than reference sites (not statistically different, Table 4). Perimeter channels had a significantly lower percentage of Ephemeroptera, Plecoptera, and Trichoptera (EPT) (5%) than reference sites (48%) and a significantly lower EPT richness (1 vs 4) (Table 4). These parameters, along with WVSCI score, were significantly different with regards to mean conductivity (Table 11). Additional ANCOVA analysis revealed significant interactions between site type and site conductivity for percent EPT, EPT richness, and total richness (Table 11, Fig. 43-46).

Macroinvertebrate data were used to rank the quality of site habitat using the West Virginia Stream Condition Index (WVSCI). WVSCI ranks overall stream quality based on measures of the benthic invertebrate community. The higher the score, the better the stream condition. Perimeter channels had an average WVSCI score of 48 (Poor) and reference sites had an average score of 68 (Marginal). Excluding R_WO, the mean score for reference sites was 74 (Good). Perimeter channels ranged from poor to marginal and reference sites ranged from poor to excellent.

Both perimeter channel and reference sites were dominated by collector-gatherer functional feeding groups (Table 15). Perimeter channels were composed of 74%

collector-gatherers, 8% predators, 8% omnivores (primarily planktonic species), and 2% shredders. Reference sites were composed of 57% collector-gatherers, 27% shredders, 5% predators, and 3% omnivores. Perimeter channel communities were composed of primarily lentic-inhabiting species while reference communities were primarily lotic-inhabiting species. Macroinvertebrate abundance data are reported in Appendices F-H.

NMDS analysis showed clustering of sites by site type with P_AR tending to have a community type more similar to reference sites and R_HC having a community type more similar to perimeter sites (Fig. 47). P_WO and R_WO were somewhat separate from the rest of the sites and plotted out in the positive NMDS 1 and positive NMDS 2 quadrant. Increasing selenium, ammonia, and summer discharge were significant predictors of community composition but did not seem to be associated with any particular group of sites. Additionally, WVSCI score, percent tolerant taxa, percent EPT, EPT richness, percent unknown feeding group, percent shredder, and percent omnivore were correlated with community composition (Fig. 48).

4.8 Organic Matter Retention

Retention was significantly higher in perimeter channels than in reference sites for measures of mean cumulative stick distance traveled, mean cumulative stick distance per day, number of sticks exiting the reach, and percent of sticks retained (Table 4, Table 16). No movement of artificial sticks or leaves was recorded in any perimeter channel sites except P_WO. Mean distance traveled by artificial leaves at P_WO was 2.60 m. Mean cumulative distance traveled per day by artificial sticks at P_WO was 0.06 m/day. Mean cumulative distance traveled by artificial sticks was 0.00 m/day in perimeter channels, excluding P_WO, and 0.06 m/day in reference sites. Mean distance traveled by artificial leaves was 0.00 m in perimeter channels, excluding P_WO, and 0.42 m in reference sites. Overall retention rate was not significantly different between site types. Perimeter channels averaged a rate of -0.064 and reference sites averaged -0.020.

4.9 Dissolved Carbon

Dissolved Organic Carbon

Perimeter channels averaged higher dissolved organic carbon (DOC) concentrations than reference sites (Table 17, Fig. 49), but only mean winter DOC levels were significantly higher in perimeter channels than reference sites (Table 4). Mean DOC for three seasons with water was 3.27 ± 2.09 mg/L for perimeter channels and 1.51 ± 0.64 mg/L for reference sites. Statistically significant trends were shown for DOC in regards to season (Table 7). Seasonal DOC was not correlated with seasonal discharge as expected (McDowell & Likens 1988c, Collier et al. 1989, Hinton et al. 1997, Meyer et al. 1998, Dawson et al. 2002, Spencer et al. 2007).

Total Carbon

Total Carbon (TC) measures were statistically higher in perimeter channels than reference sites for all seasons (Table 4, Fig. 50). Mean TC for spring, summer, and winter (reference sites did not contain enough water for sampling in autumn) was 18.5 ± 2.1 mg/L for perimeter channels and 2.6 ± 0.3 mg/L for reference sites (Table 18).

4.10 Decomposition and Processing Power

Perimeter channels averaged 47 ± 2% loss of organic matter after about 325 days, whereas reference sites averaged 62 ± 19% loss (Table 19). Reference sites lost 41%

(R_HC) (200 days), 89% (R_LF), 71% (R_ME), 63% (R_MW), and 48% (R_WO) (Fig. 51). However, when calculated, neither the decomposition rate nor the percent organic mass lost were significantly different at a 95% confidence interval. When the effects of conductivity were removed, however, mean total weight of litter bags after 325 days was significantly different between site types (Table 11).

The mean calculated decomposition rate for perimeter channels was -0.00213 ± 0.00038 and the mean rate for reference sites was -0.00348 ± 0.00196. Figure 52 shows the rates for each site. These rates are within the range of 0.0014-0.021 suggested by Beiser et al. (1991) and D'Angelo and Webster (1992). Decomposition rate was positively correlated with WVSCI score, percent EPT, percent unknown invertebrates, total number of adult amphibians, total number of adult amphibian species, mean percent of organic matter lost, and mean cumulative stick distance (Table 20). Percent shredders was not correlated with decomposition as expected (Table 20). Additionally, ANCOVA analysis revealed significant interactions between site type and site conductivity for decomposition rate, mean percent organic matter remaining, and mean percent of organic matter lost (Table 11, Fig. 53-55).

Overall processing power was not statistically different between site types (Table 4). Processing power averaged 0.013 in perimeter channels and 0.007 in reference sites (Table 19).

4.11 Structural Changes over Time

Overall, few correlations between perimeter channel characteristics and age since reclamation were observed. Chloride and sodium (Fig. 56) were correlated with age primarily because of elevated levels within the oldest site P_BH. TDS and conductivity (Fig. 57) were not correlated with age since reclamation; conductivity remains elevated over time. In terms of vegetative community structure, percent cattail was correlated with age as well as percent fern, species per km^2, and trees per km^2 (Fig. 58 & 59). However, correlations of the former three categories are due to zero values for all sites except for P_BH. Macroinvertebrate community structure showed significant correlation with age since reclamation in terms of total family richness (Fig. 60). Correlation with percent filterer was due to 2% presence in P_ST and 8% in P_BH. Percent tolerant species and percent chironomid were not correlated with age (Fig. 61). Total adult amphibian species and percent grassland amphibians were negatively correlated with age since reclamation (Fig. 62). No decomposition parameters were correlated with age since reclamation.

4.12 The Effects of Elevated Conductivity

Conductivity and associated parameters were some of the most outstanding differences between site types. ANCOVA analysis was run on decomposition and biotic metrics to determine any confounding effects of elevated conductivity levels on measures within sites. Significant interactions between site type and conductivity were found for decomposition rate, mean percent organic matter after 325 days, mean percent organic mass lost, WVSCI score, percent EPT, EPT richness, total invertebrate richness, amphibian richness, larval amphibian richness, total number of amphibians, mean number of amphibians, total number of larval amphibians, mean amphibian density, and percent forest-utilizing amphibians (Table 11). Additionally, mean total weight after 325 days, WVSCI score, percent EPT, EPT richness, amphibian richness, and larval amphibian richness were found to be significantly different by mean conductivity.

4.13 Ecological Units

EUs were calculated from parameters deemed to be important characterizers between perimeter and reference sites. Among those with means greater in perimeter channels were percentage of mean larval amphibian biomass, mean conductivity, retention rate, mean DOC, processing power, lentic amphibian species, and macroinvertebrate species richness. Those with means greater in reference sites were invertebrate biomass, WVSCI score, decomposition rate, EPA RBP, EPT richness, percentage EPT, and percentage of lotic amphibian species (Table 21).

5.0 Discussion

5.1 Key Concerns

Elevated conductivity appeared to be the primary determinant of reduced biological conditions and ecosystem processes in reclaimed perimeter channels. ANCOVA analysis revealed key interactions between site type (perimeter channel and reference site) and conductivity with regards to OM decomposition and invertebrate and amphibian metrics (Table 11).

<u>The Effect Conductivity and Site Type on Decomposition</u>

Although not previously indicated in published literature, ANCOVA analysis indicated a significant effect of site type and conductivity on OM decomposition rate (Table 11). Overall analysis indicated that conductivity may be influencing decomposition rate in perimeter channels, possibly through reduced microbial activity. Simon et al. (2009) found differences in stream OM decomposition rates along a pH gradient. They determined that reduced decomposition was the result of altered microbial assemblages and reduced microbial activity under low pH conditions (Simon et al. 2009). Additional studies have linked reduced microbial biomass and respiration with reduced decomposition rates within acidified streams (Mulholland et al. 1987, Griffith & Perry 1994, Meegan et al. 1996, Niyogi et al. 2001). Although the sites in this study did not suffer from reduced pH (except for R_WO), increased conductivity may have similar effects on microbial community structure, biomass, or respiration.

The Effect Conductivity and Site Type on Macroinvertebrates

Within the central Appalachian region, the order Ephemeroptera usually account for 25-50% of the total spring macroinvertebrate community in relatively undisturbed headwater streams (Pond et al. 2008). Ephemeroptera have also been found to show the greatest response to increases in specific conductivity in waters affected by surface mining within the region (Hartman et al. 2005, Pond et al. 2008). Consistent with previous studies, the relatively undisturbed reference sites contained a significantly higher percentage of EPT than perimeter channel sites. Additionally, a significant interaction between site type and conductivity was shown for WVSCI score, percent EPT, EPT richness, and total invertebrate richness (Table 11). EPT species are considered taxa indicative of good water quality and many cannot be supported in perimeter channel sites due to conversion from lotic to lentic conditions in addition to elevated TDS and specific conductivity (Pond et al. 2008). One possible mechanism for this is the relationship between elevated conductivity and interference with the osmoregulation of macroinvertebrates (Wichard 1973, McCulloch 1993).

The shift in community composition from a community supporting a large percentage of shredders to a community supporting a large percentage of collector gatherers (primarily chironomids) may have downstream implications. Shredders play an important role within the aquatic continuum by feeding on coarse particulate organic matter (CPOM) and converting it to fine particulate organic matter (FPOM) (Cummins & Klug 1979). FPOM, in turn, is exported as a food resource base for collector-gatherers (Short & Maslin 1977). If the shredder community is lost or reduced, shortcomings in trophic linkages may affect the entire aquatic ecosystem.

The Effect Conductivity and Site Type on Amphibians

Elevated conductivity levels also had a significant influence on amphibian assemblage composition. Specifically, a strong interaction was shown between conductivity and site type for the numbers of amphibians (primarily larva), overall richness, amphibian density, and percent of forest-utilizing species (Table 11). Most of these metrics are associated with the larval component of the amphibian population. These metrics are quantitative indicators of the shift in community compositions between site types. Differences in water chemistry such as elevated sulfate, calcium, magnesium, and alkalinity, however, did not seem to deter amphibians from using these sites overall.

The difference in number of adults versus larval amphibians at the two site types can be explained by the differences in the communities that both inhabit and use the aquatic features. Reference sites were inhabited by stream salamanders that live and breed within the stream. The difference in quantity of adults and larva at these sites may be due to the ability to more easily locate and capture the larger, adult salamanders as well as the relatively small number of eggs deposited by these salamander species (~20) (Green & Pauley 1987). The large quantity of larval versus adult amphibians in perimeter channels may be explained by the utilization of these sites by both lentic-using species as well as tree frogs inhabiting adjacent, intact forests that utilize these lentic sites for breeding. Both lentic frogs and tree frogs may lay as many as 1000-2000 eggs (Green & Pauley 1987). Frog larva are more easily captured than stream salamander larva as they tend to congregate at pond margins and are more visible. Additionally, frog adults are harder to locate and capture than their larva.

5.2 Ecological Units

To assist with quantitative calculation of on-site shortcomings, EUs have been developed. EUs present a proportional difference in measures found at perimeter channel sites versus those found at reference sites. The differences represent aspects of the original site that have been lost and would be difficult to restore on-site because of the conversion of low TDS lotic channels to high TDS lentic channels. Overall the EUs show that perimeter channels were functionally similar to reference channels in terms of amphibian biomass and OM processing. Substantial shortcomings were present with regards to species composition shifts, losses of sensitive invertebrate taxa, and overall invertebrate taxa richness. Attempts to compensate for shortcomings captured in the ratios by re-constructing stream structure on-site are ill-advised because of the difficulties associated with constructing a lotic system “from scratch” (Palmer et al. 2009).

Therefore, perimeter channels should be designed as lentic systems and EUs should be applied to sites outside the mine permit boundary on an aquatic surface area basis (Merovich & Petty 2007). For example, to compensate for a reduction in overall WVSCI score, off-site mitigation projects to enhance lotic habitat for invertebrates can be conducted at a rate of 1 meter of mitigation for every 0.71 m of perimeter channel. The application of EUs at a watershed scale may allow off-set of functional and structural losses that occur despite reclamation efforts.

5.3 Reclamation Successes

Amphibian Community

Perimeter channel and reference sites supported two very different amphibian community types, however, overall diversity and number of species supported was comparable. Perimeter channels supported a majority of generalist species, such as *Rana clamitans* (green frog) and *Notophthalmus v. viridescens* (red spotted newt), and tree frog larva such as *Hyla chrysoscelis* (Cope's gray tree frog). Reference sites supported stream salamanders, primarily *Desmognathus monticola* (seal salamander) and *Desmognathus fuscus* (northern dusky salamander).

Overall, preferences of the species themselves to use or inhabit lentic versus lotic waters were the driving factors of community composition. Primarily those species that prefer lotic habitats were found in reference sites and those species preferring lentic habitats for breeding or feeding were found in perimeter channel habitats. However, the vegetational differences between these site types may be the second most important characteristic. The amphibian community structure is highly correlated to the type and quality of vegetation present. Young, sparsely vegetated perimeter channels (such as P_WO) did not support larval amphibians despite being equally as close to intact forest as other perimeter channel sites.

In terms of number of species, both site types supported an average of two larval species and approximately two adult species. Overall diversity remained the same; however, there was an unmistakable shift in community type. Lotic and forest species were replaced by grassland-inhabiting, lentic species. Perimeter channels supported the larva of forest species such as *Hyla chrysoscelis* (Cope's gray tree frog) and *Pseudacris c. crucifer* (northern spring peeper) (Table 13, Appendix D). These sites most likely benefited from close proximity to intact forest (Hecnar & M'Closkey 1996, Stevens et al. 2002). Constructed wetlands have been reported to be colonized by ubiquitous anurans such as gray tree frog, American toad, spring peeper, and *Rana catesbeiana* (American bullfrog) within two years of creation (Perry et al. 1996, Mierzwa 2000, Pechmann et al. 2001). All of the perimeter channels in this study were older than three years since reclamation and showed colonization by amphibians. No positive correlation between amphibian community metrics and age since reclamation was found. Negative correlations were shown between total adult amphibian species and percent of grassland amphibians and age since reclamation (Table 4). This means that the number of adult amphibians, presumably grassland species, reduced over time. It is difficult to determine why this might have occurred by sampling five perimeter channels. As the perimeter channels age, there may be a significant change in an unmeasured parameter such as mean water depth or cattail density that dissuaded use by grassland adults.

The colonization by amphibians shortly after wetland establishment is consistent with patterns found in accidentally formed and constructed wetlands (Kent & Langston 2000, Pollio 2005). Although other studies found greater numbers of species in these wetlands than we found (Pollio 2005), this study found six of nine species expected to occur in grassland areas (Table 13). Additionally, studies have shown the number of amphibian species found in newly created pools was positively correlated with the distance of these pools to forests (Laan & Verboom 1990). As the perimeter channels in this study were not intentionally designed as wetlands to support amphibians, considering wetland habitat during perimeter channel construction may lead to an increase in the number of species using the wetlands, as long as minimal distance to intact forest is maintained.

Macroinvertebrate Community

Perimeter channels and reference sites had similar macroinvertebrate family richness and biomass but significantly different community composition. Perimeter channels overall, however, supported a comparable number of macroinvertebrates and number of species. Perimeter channels were dominated by lentic species, such as Odonates, and were dominated by collector-gathers (primarily chironomids). Reference sites were dominated by lotic species and had a higher percentage of shredders than perimeter channel sites.

Decomposition and Carbon Cycling

Mean decomposition rates for both site types were within ranges suggested by Beiser et al. (1991) and D'Angelo and Webster (1992). OM decomposition rates showed a significant overall effect of site type and mean conductivity. Decomposition rates were not, however, correlated with parameters known to affect decomposition such as temperature, nutrient concentration, hydrology, dissolved oxygen, percentage of shredders, or pH (Whiles & Wallace 1997, Graça et al. 2001, Swan 2004, Simon et al. 2009) (Table 4). However, these parameters were also not significantly different between site types with the exception of summer pH and spring discharge. Although discharge did not differ between site types, hydrology in terms of lentic versus lotic systems did. Therefore, it is likely that, the difference in aquatic system type itself affects the potential breakdown rate between perimeter channel and reference site types.

Field observations over the course of the study support the idea that the lotic component to sites contributed to their overall loss of organic matter. The position of the bags and securing rope in relation to the rebar stake, the amount of movement from the original placement, the degree of distress to the mesh bag, and the integrity of the remaining material indicate that the majority of material lost from reference site bags was lost through mechanical abrasion.

The difference in the degree of water flow between site types is evidenced by differences in organic matter retention. No movement of artificial sticks or leaves was recorded at any perimeter channel sites except P_WO (which has the highest gradient of perimeter channels) (Table 16). Of the perimeter channel sites, P_WO also had the highest decomposition rate (Table 19). Overall distance traveled by artificial sticks and leaves was greater in reference sites than in perimeter channels.

Winter DOC levels in perimeter channels were statistically higher than in reference sites, despite a loss of original topsoil. Because of the relation between DOC and soil type and chemistry (McDowell & Likens 1988, Dawson et al. 2002, Ankers et al. 2003), it is unlikely that the DOC in perimeter channel systems originates from the soil of the reclaimed site. Perimeter channel DOC is also unlikely to originate from leaf litter inputs as found by Hongve (1999). Perimeter channels have a low percent canopy cover and are usually at or above the canopy height of adjacent intact forest.

This knowledge, along with comparable processing power rates between site types, indicates that the high DOC concentrations within perimeter channels is most likely originating from on-site inputs of detritus from aquatic macrophytes. Specifically, the high percent cover emergent vegetation and higher retention capability of perimeter channels allows organic material to be effectively cycled within the perimeter channels Anderson and Mitsch (2006) found that the percentage of soil organic matter content within riverine wetlands increased about one percent every three years. This concurs with correlations in this study between age since reclamation and percent cattail (Table 4). The high retention ability of these sites combined with increasing cattail coverage may lead to increased mean DOC concentrations with age since reclamation. This trend is seen in all but the oldest site (P_BH) (Table 17).

Perimeter channels overall resemble wetlands more than streams. Because wetlands of this type are uncommon within West Virginia, it may be difficult to evaluate their function as wetlands. An expected range of DOC levels, especially, may be difficult to evaluate without local reference wetlands because of the complexity of parameters that determine DOC concentration as well as the natural DOC flux inherent from environmental conditions. For instance, Mann and Wetzel (1995) found DOC levels fluctuated naturally with seasonal macrophyte growth and bacterial production within riverine wetlands. Additionally, geographic region, elevation, season, and degree of exposure may influence local DOC fluctuation.

Seasonal DOC concentration was not correlated with seasonal discharge as expected (McDowell & Likens 1988c, Collier et al. 1989, Hinton et al. 1997, Meyer et al. 1998, Dawson et al. 2002, Spencer et al. 2007). This may be due to the mining disturbance at perimeter channel sites resulting in large, reclaimed catchments with relatively low OM inputs from tree canopies. Reference site concentrations were within the range of 0.673 – 2.94 mg/L for forested watersheds within the region (Meyer & Tate 1983; Tate & Meyer 1983). Perimeter channel mean concentrations were less than annual mean concentrations of 9.8 ± 1.5 mg/L for wetland-dominated watersheds reported by Eimers et al. (2008). Both mean concentrations, however, are comparable to mean annual concentrations ranging from 7.1 – 48.2 mg/L within the ponded portion of riverine wetlands (Mann & Wetzel 1995).

5.4 Are Ecological Functions Reclaimed Locally?

The process of site reclamation is intended to offset any geomorphic or ecological losses (Bradshaw 1984, Holl 2002). Although full re-creation of the original topography of an area during reclamation is considered the geomorphic ideal, it is not always appropriate, or feasible, in steeper areas (Nicolau 2003). Ecologically, Hilderbrand et al. (2005) urge the setting of realistic restoration goals and argue that “scientifically defensible end points of functional or structural equivalence" need to be set. In the case of mountaintop removal mining in West Virginia, the site is converted from a steep, forested headwater stream to an unforested site with rolling terrain and wetland-like aquatic features. Despite this conversion, the question remains if ecological functions such as supporting biological communities and downstream energy export are adequately reclaimed.

Functional downstream export of carbon is supported by the on-site generation of DOC. Higher retention capabilities of perimeter sites allows for increased opportunity for decomposition. Overall processing power is comparable in perimeter sites and reference sites. Mechanical breakdown of OM is lost, but decomposition rates are comparable.

In terms of biotic communities, both amphibians and macroinvertebrates showed similar diversities on mined and unmined sites. However, communities shifted from lotic communities supporting sensitive taxa to lentic communities supporting generalists and tolerant taxa. Any retention of biotic communities by perimeter channels may be favorably influenced by the proximity of intact forest. These areas may encourage biotic use of perimeter sites and act as a source for colonizers.

5.5 Watershed Scale Perspective

The change in site type from forested stream to perimeter channel resulted in a shift in vegetation, amphibian, and macroinvertebrate composition that cannot be reclaimed on-site. The new system cannot support the same community types. In response to the loss of pre-existing communities, off-site mitigation that supports healthy, native biotic communities must be pursued.

Additionally, the shift in communities from lotic, sensitive taxa to lentic, generalist taxa may become problematic as the cumulative effects from stream to stream and watershed to watershed are considered over a region (Lowe & Bolger 2002, Lowe et al. 2006, Pond et al. 2008). Disturbance at a local scale may influence the ability of populations to re-colonize at a regional scale (Lowe & Bolger 2002). Although prevention of species loss at a local scale may not be possible, it can be prevented at the watershed scale. Consideration must be given to protect portions of a mined watershed to act as sites for wildlife protection and source populations for continued re-colonization (Lowe et al. 2006, Pond et al. 2008). Consideration must be given both to the extent of the watershed affected and to the life histories of at-risk species to determine regional habitat needs (Lowe et al. 2006). Ensuring the connectivity of first-order streams may be essential for ensuring the survival of some species, such as *Gyrinophilus porphyriticus* (spring salamander) (Lowe & Bolger 2002, Lowe et al. 2006).

The change in water chemistry resulting in increased alkalinity, manganese, TDS, calcium, magnesium, sulfate, and specific conductivity also cannot be reclaimed on-site. This increase in water parameters can be a compounded problem in the watershed as a whole depending on the extent to which the watershed is mined. Pond et al. (2008) found that the evidence of mining and reclamation on the water chemistry (especially specific conductivity) was greatly reduced in watersheds that contained a higher percentage of unmined tributaries. The best way to handle changes in water chemistry resulting from watershed scale disturbance may be dilution at a watershed scale, primarily through planned protection of headwater streams (Saunders et al. 2002. Lowe et al. 2006). By preserving a percentage of tributaries within the mined watershed as undisturbed sites and sources of dilute water, cumulative downstream changes in water chemistry may be avoided (Saunders et al. 2002, Merriam 2009). This may be an especially pertinent solution because mining and its effects may not be the only stressor to local watersheds within the region. Historical mining and non-residential development may compound watershed-wide stresses to ecological function (Merriam 2009).

5.6 Reference Site Condition

Reference sites were selected for this study based on winter and spring water chemistry measures, topographic maps, and a general knowledge of the area. The reference streams selected were known to drain watersheds with no surface mining activities and no residential development. Preliminary measures of water quality indicated that all streams were in reasonably good condition for streams in this region.

Unfortunately, two of the reference sites, R_HC and R_WO, later displayed water chemistry that was less than ideal. R_WO had lower than average pH as well as higher acidity, aluminum, manganese, calcium, manganese, conductivity, and sulfate than other reference sites. The water chemistry of this site is indicative of streams impacted by acid mine drainage (Merovich et al. 2007). Consequently, this site is presumed to be influenced by historic underground mining. It is not clear why initial water quality measures did not show signs of impairment. However, we know from studies in other watersheds that water quality in streams impacted by acid mine drainage (AMD) can vary significantly from one season to the next (Merovich et al. 2007). The poor water quality conditions of R_WO likely contributed to measures of organic matter decomposition, and amphibian and macroinvertebrate diversity that were substantially lower than those of other reference sites.

Another reference site, R_HC, was impacted by some unknown, non-mining related disturbance upstream in summer 2008. During this period, a natural gas line and access road were installed and waste water from gas well drilling may have been introduced to the stream. Prior to disturbance, R_HC possessed water quality characteristics very similar to the other high quality reference sites. Following disturbance, all pore spaces within the R_HC stream bed were filled with sediment resulting in loss of habitat for amphibians and macroinvertebrates. Additionally, R_HC water chemistry measures showed an increase in nitrite, conductivity, barium, sodium, and chloride.

Although R_HC and R_WO possessed less-than-ideal biotic and abiotic conditions, these sites were included in most analyses comparing un-mined reference stream channels to reclaimed mine perimeter channels. Excluding them would have reduced our sample size from five to three and made direct comparisons between site types difficult. In addition, we believe that the range of conditions observed at reference sites is representative of streams draining watersheds that have not been surface mined (Minter 2009, Merriam 2009). Because of the topography and geology of the area, it is likely that one in five watersheds in the region will be affected by legacy impacts from underground mining and increasingly streams are being impacted by gas extraction or other non-mining related disturbance (Merriam 2009).

5.7 Additional Questions

Previous literature has emphasized the connectivity of upstream functional and ecological processes to downstream ecosystem function and value (Vannote et al. 1980, Gomi et al. 2002, Lowe et al. 2006, Meyer et al. 2007, Wipfli et al. 2007). If downstream functions and values are to be maintained, the watershed function as a whole must be considered (Lowe & Bolger 2002, Saunders et al. 2002, Lowe et al. 2006, Pond et al. 2008). The functions considered by this study were the downstream export of energy in terms of on-site OM retention and decomposition and off-site DOC export and the support of biologic community structure and diversity. Additional studies may seek to understand possible losses in function of the downstream export of particulate organic matter (POM), emphasized by Vannote et al. (1980), and gaps in ecological function originating from the physical gap between perimeter channel off-site outflow points and native stream channels.

Native channels act as a source of coarse and fine particulate organic matter (CPOM/FPOM) for downstream trophic webs (Cummins & Klug 1979, Vannote et al. 1980, Cummins et al. 1989, Wallace et al. 1997). This is generated by macroinvertebrate activity as well as mechanical breakdown of organic matter. Since perimeter channels have a different macroinvertebrates community than reference sites, and because the mechanical component caused by lotic waters is lost, there may be an additional functional loss of CPOM/FPOM export. Additional studies may investigate potential shortcomings in CPOM and FPOM production and export.

The reclaimed perimeter channels in this study consisted of retained water prevented from escaping off the mine perimeter by berms. These features drained towards a central point where an off-site flow was created by perforating the berm. The water was then allowed to drain off-site, downhill and rejoin native streams. The physical area in between the point where water exits the reclaimed site and rejoins native channels may act as an additional site of disturbance as no native channel exists and exported water creates a new channel. Future studies may seek to measure the differences in water chemistry between the uphill off-site flow point and the junction where the flow joins native channels. Specifically, what changes in DOC may occur between those two points? Additionally, does the transitional zone allow for increases or decreases in TDS, total suspended solids (TSS), and specific conductivity?

The reclaimed perimeter channels in this study resemble wetlands. However, no comparison was made between the ecological function of these sites as wetlands and the ecological function of similar wetlands within the region. Rough comparisons can be made via published literature. Overall, DOC concentrations and decomposition rates were comparable to published values (Beiser et al. 1991, D'Angelo & Webster 1992, Mann & Wetzel 1995). The DOC concentrations and decomposition rates expected for wetlands within the study site may differ, however, because of regional factors such as elevation, growing season, geology, etc. Future studies may seek to compare perimeter site function to the function of comparable wetlands within the region.

This study also did not investigate differences in the functions of perimeter channels in terms of their overall construction and design. Comparison to native wetlands may aid in guiding design suggestions such as recommended percent of open water and water depth. At this point, variation exists from site to site in terms of how mine sites are reclaimed and how perimeter channels are designed. These differences seem to originate from both the time period of the reclamation and the company performing the reclamation. Future studies may seek to discover if increases in functional recovery can be gained simply by perimeter channel design. Specifically, can conductivity and TDS be further reduced on-site and can intentional design increase the diversity and structure of biotic communities?

5.8 Summary

Although perimeter channels support on-site biotic communities, decomposition rate, and processing power comparable to pre-mining condition, the manner of conducting these ecological functions differs. Perimeter channels have lost lotic components that aid in OM decomposition and downstream transport. Biotic communities have shifted from supporting sensitive, lotic taxa to supporting generalist, lentic taxa. Increases in water chemistry parameters such as alkalinity, conductivity, and related parameters cannot be mitigated for on-site and must be considered at a watershed scale. Native headwater streams within the affected watershed must be protected to act as refugia and source populations for biota and sources of fresh-water for the watershed-wide dilution of exported water. Off-site mitigation can compensate for on-site shortcomings through the application of EUs. Additional research may provide insights into the linkages between perimeter channels and native catchments and suggestions for future perimeter channel design.

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Tables

Table 1. Site name, site code, site type, approximate age, mean discharge, calculated drainage area (DA), and HUC 10 watershed (as defined by the Natural Resources Conservation Service), latitude and longitude in decimal degrees for reclaimed mine perimeter channels and reference streams. P = reclaimed mine perimeter channel. R = reference site. na = not applicable

Site Names	Site Code	Site Type	Approx. Age (years)	Mean Discharge (m^3/s)	Calculated DA (ha)	(HUC 10) Watershed	Latitude (DD)	Longitude (DD)
White Oak	P_WO	P	3	0.01588	45	Coal River	38.04778	-81.52139
Argus	P_AR	P	5	0.00127	6	East Fork Twelvepole	37.98972	-82.25222
Stanley Branch	P_ST	P	10	0.01165	33	Mud River	38.08306	-81.93472
Sugartree	P_SU	P	10	0.01304	37	Mud River	38.09007	-81.95751
Big Horse	P_BH	P	20	0.00300	12	Little Coal River	38.08500	-81.89750
Unnamed Tributary to Hell Creek	R_HC	R	na	0.00048	7	Pigeon Creek	37.73044	-82.23232
Unnamed Tributary to Lukey Fork	R_LF	R	na	0.00280	10	Mud River	38.05944	-81.95306
Unnamed Tributary to Mud Creek East	R_ME	R	na	0.00107	5	Mud River	38.04647	-81.91148
Unnamed Tributary to Mud Creek West	R_MW	R	na	0.00287	11	Mud River	38.06105	-81.94331
Unnamed Tributary to White Oak	R_WO	R	na	0.00440	16	Coal River	38.05250	-81.52278

Table 2. Sampled parameters for reclaimed mine perimeter channels and reference sites with sampling dates.

Response Variables	Sample Dates
Mean Daily Water Temp (°C)	June 08 - June 09
Mean Q (m^3/s)	Feb 08, Mar 08, May, 08, June 08, July 08, Oct 08, Dec 08, Feb 09, May 09
Water Chemistry	Feb 08, Mar 08, June 08, Oct 08, Feb 09, May 09
Mean Alkalinity (mg/L)	Feb 08, Mar 08, June 08, Oct 08, Feb 09, May 09
Mean Acidity (mg/L)	Feb 08, Mar 08, June 08, Oct 08, Feb 09, May 09
Seasonal Water Chemistry	
Mean pH	Feb 08, Mar 08, May, 08, June 08, July 08, Oct 08, Dec 08, Feb 09, May 09
Mean Cond (µS/cm)	Feb 08, Mar 08, May, 08, June 08, July 08, Oct 08, Dec 08, Feb 09, May 09
Mean DO (mg/L)	Feb 08, Mar 08, May, 08, June 08, July 08, Oct 08, Dec 08, Feb 09, May 09
Mean TDS (mg/L)	Feb 08, Mar 08, May, 08, June 08, July 08, Oct 08, Dec 08, Feb 09, May 09
Habitat Assessment	
EPA RBP	March 08
VA USM	June 08
WV FCU	June 08
BEHI	June 08
ORAM	Oct 08
Vegetation	June 08
Macroinvertebrates	
WVSCI Score	May 08
Pct Chironomid	May 08
Pct Tolerant	May 08
Pct EPT	May 08

Table 2 continued.

Response Variables	Sample Dates
EPT Richness	May 08
Total Invertebrate Richness	May 08
Total Inverts	May 08
Total Invert Biomass (g/m^2)	May 08
Amphibians	
Amph Species Richness	Mar 08, May 08, June 08, July 08
Larval Amph Species Richness	Mar 08, May 08, June 08, July 08
Adult Amph Species Richness	Mar 08, May 08, June 08, July 08
Total No Amph	Mar 08, May 08, June 08, July 08
Total No Adult Amph	Mar 08, May 08, June 08, July 08
Total No Larval Amph	Mar 08, May 08, June 08, July 08
Mean Larval Amph Biomass ($100g/m^2$)	Mar 08, May 08, June 08, July 08
Mean Amph Density (ind/m^2)	Mar 08, May 08, June 08, July 08
Pct Grassland Amph	Mar 08, May 08, June 08, July 08
Grassland Amph Species	Mar 08, May 08, June 08, July 08
Pct Forest Amph	Mar 08, May 08, June 08, July 08
Pct Lotic Amph	Mar 08, May 08, June 08, July 08
Pct Lentic Amph	Mar 08, May 08, June 08, July 08
Lentic Amph Species	Mar 08, May 08, June 08, July 08
OM Processing	
Decomp Rate	May 08, June 08, July 08, Oct 08, Feb 09
OM Retention	
Mean Leaf Distance (m)	March 08, June 08, Oct, 08, Feb 09
Mean Cumm Stick Distance (m)	March 08, May 08, June 08, Oct 08
Retention Rate	derived
Anl Mean DOC (mg/L)	May 08, Jun 08, Oct 08, Feb 09
Processing Power * 100	derived

Table 3. Habitat assessment scores for reclaimed mine perimeter channels and reference streams. RBP= Rapid Bioassessment Protocol, USM= Unified Stream Method, FCU= Functional Channel Unit Assessment, BEHI= Bank Erosion Hazard Index, and ORAM = Ohio Rapid Assessment Method. Mean and standard deviation by site type are given in the last two rows. Perimeter channel sites are listed in increasing age since reclamation. Statistical significance is indicated by *= <0.05.

Site Code	EPA RBP*	VA USM*	WV FCU*	BEHI*	BEHI Qualifier	ORAM Score*
P_WO	55	5	3	36	High	18
P_AR	88	3	3	12	Low	21
P_ST	79	4	2	24	Moderate	48
P_SU	85	4	4	24	Moderate	44
P_BH	84	4	3	20	Low	43
R_HC	172	7	9	46	Very High	50
R_LF	132	6	8	39	High	68
R_ME	136	6	9	42	Very High	71
R_MW	133	6	10	36	High	58
R_WO	175	7	9	31	High	59
Perimeter	78 ± 13	4 ± 1	3 ± 1	23 ± 9	-	35 ± 14
Reference	150 ± 22	6 ± 0	9 ± 1	39 ± 6	-	61 ± 8

Table 4. Mean and standard error for site types. Water chemistry means were calculated using ½ the method detection limit (MDL) for values at MDL. T test statistics for means *= <0.05, **= <0.005, ***= <0.001.

Response Variables	Perimeter Mean	Reference Mean	T test P vs R (df=8)
Temperature			
Max Daily Temp (°C)	30.7 (3.2)	31.9 (2.4)	-0.2922
Min Daily Temp (°C)	0.15 (0.08)	0.00 (0.00)	2.2826
Mean Daily Temp (°C)	10.5 (0.2)	9.6 (0.7)	1.1182
CV for Mean Daily Temp	63.6 (3.5)	58.9 (4.8)	0.7953
Discharge			
Mean Spring Q (m^3/s)	0.011 (0.004)	0.001 (0.001)	2.7123*
Mean Sum Q (m^3/s)	0.003 (0.003)	0.000 (0.000)	1.0716
Mean Aut Q (m^3/s)	0.006 (0.004)	0.001 (0.001)	1.1153
Mean Winter Q (m^3/s)	0.017 (0.005)	0.006 (0.002)	1.8001
Mean Q (m^3/s)	0.009 (0.003)	0.002 (0.001)	2.1576
Water Chemistry			
Mean Alkalinity (mg/L)	138 (13)	5 (1)	12.8148***
Mean Acidity (mg/L)	0 (0)	13 (5)	-4.8816**
Mean Al (mg/L)	0.1 (0.0)	0.5 (0.5)	-0.867
Mean Ca (mg/L)	163 (31)	21 (15)	4.9602**
Mean Fe (mg/L)	0.10 (0.04)	0.06 (0.00)	1.0342
Mean Mg (mg/L)	154 (34)	9 (4)	6.1238***
Mean Mn (mg/L)	0.3 (0.2)	0.4 (0.3)	0.2048
Mean SO_4 (mg/L)	1008 (196)	32 (20)	7.7072***
Mean Cl (mg/L)	13.8 (9.7)	11.4 (10.2)	0.4478
Mean NO_2 (mg/L)	0.04 (0.03)	3.10 (3.08)	-0.9352
Mean NO_3 (mg/L)	11.0 (10.4)	0.8 (0.2)	0.8778
Mean TP (mg/L)	0.08 (0.04)	0.05 (0.01)	0.6656
Mean NH_3 (mg/L)	0.007 (0.003)	0.013 (0.002)	-1.9669
Mean Ba (mg/L)	0.016 (0.002)	0.093 (0.060)	-1.7213

Table 4 continued.

Response Variables	Perimeter Mean	Reference Mean	T test P vs R (df=8)
Mean Cd (mg/L)	0.007 (0.000)	0.008 (0.000)	-0.6325
Mean Co (mg/L)	0.011 (0.002)	0.009 (0.001)	1.0018
Mean Cr (mg/L)	0.006 (0.000)	0.009 (0.001)	-2.5058*
Mean Cu (mg/L)	0.008 (0.000)	0.008 (0.000)	NA
Mean Na (mg/L)	13.2 (6.3)	23.7 (22.2)	0.0937
Mean Ni (mg/L)	0.034 (0.019)	0.024 (0.011)	0.4772
Mean Zn (mg/L)	0.020 (0.008)	0.042 (0.031)	-0.4481
Mean Se (mg/L)	0.024 (0.002)	0.030 (0.005)	-1.019
Mean Spring pH	7.3 (0.1)	6.9 (0.5)	0.7736
Mean Summer pH	7.2 (0.2)	5.7 (0.3)	4.0200**
Mean Autumn pH	7.7 (0.1)	7.1 (0.5)	1.4153
Mean Winter pH	7.4 (0.1)	7.3 (0.5)	0.1104
Mean pH	7.4 (0.1)	6.7 (0.4)	1.5589
Mean Spr Cond (µS/cm)	2310 (495)	602 (445)	2.5668*
Mean Sum Cond (µS/cm)	2255 (488)	642 (501)	2.3055
Mean Aut Cond (µS/cm)	2147 (448)	133 (75)	4.5142**
Mean Win Cond (µS/cm)	2077 (395)	147 (64)	4.8184**
Mean Cond (µS/cm)	2197 (414)	461 (326)	3.2938*
Mean Spring DO (mg/L)	6.8 (0.6)	7.7 (1.0)	-0.7069
Mean Sum DO (mg/L)	9.1 (1.0)	10.4 (2.9)	-0.2203
Mean Aut DO (mg/L)	9.5 (1.3)	10.2 (1.3)	0.5170
Mean Winter DO (mg/L)	9.3 (1.1)	11.7 (0.7)	-1.7323
Mean DO (mg/L)	8.7 (0.5)	9.9 (1.0)	-1.1108
Spring TDS (mg/L)	1366 (294)	76 (28)	5.9862***
Summer TDS (mg/L)	1818 (455)	157 (95)	4.5826**
Autumn TDS (mg/L)	-	-	-

Table 4 continued.

Response Variables	Perimeter Mean	Reference Mean	T test P vs R (df=8)
Winter TDS (mg/L)	27.0 (5.0)	3.7 (0.7)	6.8737***
Mean TDS (mg/L)	25.2 (3.0)	2.7 (0.4)	6.8135***
Spring TC (mg/L)	3.3 (0.6)	-	6.1920***
Summer TC (mg/L)	3.4 (0.6)	1.4 (0.1)	10.3569***
Autumn TC (mg/L)	15.6 (2.0)	2.6 (0.3)	-
Winter TC (mg/L)	18.5 (2.1)	2.6 (0.3)	3.4641*
Mean TC (mg/L)	27.0 (5.0)	3.7 (0.7)	9.6959***
Habitat Assessment			
EPA RBP	78 (6)	150 (10)	6.2240***
VA USM	4 (0)	6 (0)	-5.3492***
WV FCU	3 (0)	9 (0)	-14.6677***
BEHI	23 (4)	39 (3)	-3.2882*
ORAM	35 (6)	61 (4)	-3.5899*
Vegetation			
Pct Bare Ground	3 (2)	29 (8)	-3.2254*
Pct Cattail	21 (7)	0 (0)	3.0614*
Pct Fern	0 (0)	11 (2)	-5.1953***
Pct Forb	22 (8)	22 (5)	0.0269
Pct Grass	22 (4)	2 (1)	5.5151***
Pct Open Water	13 (6)	0 (0)	2.0459
Pct Tree	1 (1)	16 (3)	-4.1779**
Pct Vine	2 (1)	8 (2)	-3.1965*
Species per km^2	0.1 (0.1)	2.5 (0.3)	-9.0068***
Trees per km^2	0.3 (0.3)	9.2 (2.2)	-6.3294***
Pct Canopy Cover	4 (4)	91 (1)	-26.1052***

Table 4 continued.

Response Variables	Perimeter Mean	Reference Mean	T test P vs R (df=8)
Macroinvertebrates			
WVSCI Score	48 (5)	68 (8)	-2.0154
Pct Chironomid	58 (16)	32 (11)	1.4183
Pct Tolerant	70 (15)	42 (13)	1.5593
Pct EPT	5 (4)	48 (16)	-2.4847*
EPT Richness	1 (0)	4 (1)	-2.6662*
Total Richness	8 (2)	7 (1)	0.5282
Total Inverts	763 (273)	213 (87)	1.8116
Pct 2 Dominant Sp	78 (10)	66 (6)	1.2398
Total Biomass (g/m^2)	31.8 (18.9)	34.5 (14.9)	-0.0852
Pct Collector- Gatherer	74 (12)	57 (9)	1.3313
Pct Filterer	2 (2)	0 (0)	1.3208
Pct Scraper	6 (4)	3 (3)	0.5337
Pct Shredder	2 (2)	27 (11)	-2.1756
Pct Predator	8 (3)	5 (3)	0.8231
Pct Omnivore	8 (5)	3 (3)	0.7782
Pct Unknown	0 (0)	5 (2)	-2.5767*
Amphibians			
Total No Amph Sp	3 (1)	3 (0)	-0.6040
Total Larval Amph Sp	2 (1)	2 (0)	0.1989
Total Adult Amph Sp	1 (0)	2 (0)	-2.1909
Total No Amph	24 (14)	36 (14)	-0.6352
Mean No Amph	6 (4)	9 (4)	-0.6352
Total No Adult Amph	5 (3)	28 (13)	-1.9154

Table 4 continued.

Response Variables	Perimeter Mean	Reference Mean	T test P vs R (df=8)
Total No Adult Amph	5 (3)	28 (13)	-1.9154
Mean No Adult Amph	1 (1)	7 (3)	-2.0744
Total No Larval Amph	19 (11)	8 (3)	0.7208
Mean No Larval Amph	5 (3)	2 (1)	2.0714
Total Larval Amph Biomass (100g/m^2)	0.053 (0.041)	0.002 (0.001)	1.2522
Mean Larval Amph Biomass (100g/m^2)	0.0133 (0.0103)	0.0005 (0.0002)	1.2522
Mean Amph Density (ind/m^2)	1.2 (0.7)	1.8 (0.7)	-0.6386
Pct Grassland Amph	58 (18)	0 (0)	2.7647*
Pct Forest Amph	95 (5)	100 (0)	-1.0000
Pct Lotic Amph	5 (3)	86 (13)	-5.1102***
Pct Lentic Amph	89 (8)	54 (8)	3.0617*
Decomposition			
Decomp Rate	0.0021 (0.0002)	0.0035 (0.0009)	-1.5186
Mean Total Wt (g) (325 d)	6.7 (0.9)	5.3 (1.6)	0.7314
Mean % Organic (325 d)	85 (7)	78 (6)	0.5694
Mean Organic Mass (g) (325 d)	5.3 (0.1)	3.8 (0.9)	1.7560
Mean % Org Mass Lost (325 d)	47 (1)	62 (9)	-1.7389
OM Retention			
Mean Leaf Distance (m)	0.52 (0.52)	0.42 (0.15)	-0.6724
Mean Cum Stick Distance (m)	7.44 (7.44)	26.89 (4.81)	-2.9434*
Mean Cum Stick Dist/ Day	0.04 (0.04)	0.14 (0.02)	-2.9434*
Retention Rate	-0.064 (0.014)	-0.020 (0.005)	2.0714
Pct Sticks Retained	87 (13)	59 (9)	2.5499*
No Sticks Exiting Reach	6 (6)	21 (5)	-3.7641*

Table 4 continued.

Response Variables	Perimeter Mean	Reference Mean	T test P vs R (df=8)
Dissolved Organic Carbon			
Spring DOC (mg/L)	4.9 (1.7)	2.5 (0.6)	1.0170
Summer DOC (mg/L)	2.8 (1.2)	1.0 (0.2)	1.6977
Autumn DOC (mg/L)	4.2 (1.4)	-	-
Winter DOC (mg/L)	2.1 (0.4)	1.0 (0.1)	3.0508*
Mean DOC (mg/L)	3.5 (0.9)	1.5 (0.3)	2.0183
Processing Power			
Processing Power * 100	0.013 (0.003)	0.007 (0.002)	1.6318

Table 5. Percentage of vegetation from vegetation survey for reference sites and reclaimed mine perimeter channels. Mean and standard deviation by site type are given in the last two rows. Perimeter channel sites are listed in increasing age since reclamation. Statistical significance is indicated by *= <0.05.

Site Code	Bare Ground*	Cattail*	Fern*	Forb	Grass*	Open Water	Tree*	Vine*
P_WO	11	3	0	13	19	38	5	6
P_AR	0	7	0	51	30	4	0	0
P_ST	0	34	0	8	31	8	0	3
P_SU	2	24	0	27	21	8	0	0
P_BH	3	34	2	10	11	7	2	0
R_HC	23	0	16	31	2	0	16	5
R_LF	40	0	8	11	2	0	11	11
R_ME	51	0	7	7	0	0	9	9
R_MW	26	0	8	29	5	0	14	5
R_WO	6	0	15	31	0	0	28	12
Perimeter	3 ± 5	21 ± 15	0 ± 1	22 ± 18	22 ± 8	13 ± 14	1 ± 2	2 ± 3
Reference	29 ± 17	0 ± 0	11 ± 5	22 ± 12	2 ± 2	0 ± 0	16 ± 7	8 ± 4

Table 6. Percent canopy cover and tree count data for reference sites and reclaimed mine perimeter channels. Mean and standard deviation by site type are given in the last two rows. Perimeter channel sites are listed in increasing age since reclamation. Statistical significance is indicated by *= <0.05.

Site Code	Species* per $1000m^2$	Trees* per $1000m^2$	Pct* Canopy Cover
P_WO	0.0	0.0	0
P_AR	0.0	0.0	1
P_ST	0.0	0.0	20
P_SU	0.0	0.0	0
P_BH	0.4	1.3	1
R_HC	2.1	3.8	89
R_LF	1.7	9.2	93
R_ME	2.9	10.4	92
R_MW	2.5	5.8	92
R_WO	3.3	16.7	90
Perimeter	0.1 ± 0.2	0.3 ± 0.6	4 ± 9
Reference	2.5 ± 0.7	9.2 ± 5.0	91 ± 2

Table 7. Repeated measures ANOVA analysis of seasonal parameters for reclaimed mine perimeter channels and reference streams. Statistical significance is indicated by *= <0.05, **= <0.005, ***= <0.001.

Parameter	Type df = 1	Season df = 3	Type:Season df = 3
pH	4.1082	1.2056	1.4659
Temp (°C)	6.1466*	24.2531***	4.1556*
Cond (µS/cm)	14.486**	1.1711	0.3070
DO (mg/L)	0.0646	19.6173***	0.3648
TDS (mg/L)	24.343**	1.5243	0.7794
Q (m^3/s)	5.0276	12.071***	1.0351
DOC (mg/L)	3.0713	5.4466*	0.0508

Table 8. Temperature data (°C) for reference sites and reclaimed mine perimeter channels for periods when streams contained water. Mean and standard deviation by site type are given in the last two rows. Perimeter channel sites are listed in increasing age since reclamation. No data were recovered from P_WO. Statistical significance is indicated by *= <0.05, **= <0.005, ***= <0.001.

Site Code	Max Daily Temp	Min Daily Temp	Mean Daily Temp	CV for Mean Daily Temp	Mean Daily Temp Range
P_WO	-	-	-	-	-
P_AR	22.08	0.41	9.82	54.51	1.54 ± 1.27
P_ST	26.45	0.14	10.89	63.20	3.85 ± 2.70
P_SU	40.57	0.00	10.93	63.96	4.45 ± 2.48
P_BH	34.57	0.00	10.10	75.97	6.03 ± 4.66
R_HC	38.90	0.00	12.51	42.44	4.89 ± 6.28
R_LF	31.38	0.00	9.01	69.39	5.21 ± 4.18
R_ME	23.86	0.00	9.11	54.57	3.13 ± 2.23
R_MW	34.07	0.00	8.26	62.55	4.84 ± 4.80
R_WO	31.05	0.00	9.05	65.66	5.96 ± 4.01
Perimeter	30.92 ± 8.26	0.14 ± 0.19	10.44 ± 0.56	64.41 ± 8.82	-
Reference	31.85 ± 5.46	0.00 ± 0.00	9.59 ± 1.67	58.92 ± 10.71	-

Table 9. Amphibian abundance survey totals, for four sample periods, observed on reclaimed mine perimeter channels and reference sites. Frog and salamander species' preference for grassland or forest was based on information from Green and Pauley (1987). Mean and standard deviation by site type are given in the last two rows. Perimeter channel sites are listed in increasing age since reclamation. Statistical significance is indicated by *= <0.05.

Site Code	Total Larva*	Larval Species*	Total Adults	Adult Species	Total Individuals*	Total Species*	Pct Grassland Amphibians*	Pct Forest Amphibians*	Pct Lotic Amphibians*	Pct Lentic Amphibians*
P_WO	0	0	6	2	6	2	100	100	17	100
P_AR	63	5	17	2	80	5	89	100	0	81
P_ST	9	1	3	1	12	1	25	100	8	100
P_SU	12	4	1	1	13	4	69	77	0	62
P_BH	12	2	0	0	12	2	8	100	0	100
R_HC	3	1	8	2	11	2	0	100	100	55
R_LF	8	2	45	4	53	5	0	100	98	57
R_ME	13	2	69	2	82	3	0	100	100	44
R_MW	14	2	15	2	29	3	0	100	100	83
R_WO	1	1	2	2	3	3	0	100	33	33
Perimeter	19 ± 25	2 ± 2	5 ± 7	1 ± 1	25 ± 31	3 ± 2	58 ± 40	95 ± 10	5 ± 7	89 ± 17
Reference	8 ± 6	2 ± 1	28 ± 28	2 ± 1	36 ± 32	3 ± 1	0 ± 0	100 ± 0	86 ± 30	54 ± 18

Table 10. Larval amphibian biomass (g/100m^2) for four sampling occasions observed on reclaimed mine perimeter channels and in reference sites. Mean and standard deviation by site type are given in the last two rows. Perimeter channel sites are listed in increasing age since reclamation.

Site Code	March	May	June	July	Total	Mean
P_WO	0.00	0.00	0.00	0.00	0.00	0.00
P_AR	6.91	2.91	8.22	3.37	21.41	5.35
P_ST	0.00	0.06	0.00	0.00	0.06	0.01
P_SU	0.00	0.01	0.10	0.09	0.20	0.05
P_BH	0.00	4.87	0.11	0.00	4.99	1.25
R_HC	0.00	0.06	0.00	0.00	0.06	0.02
R_LF	0.05	0.14	0.02	0.00	0.21	0.05
R_ME	0.06	0.10	0.08	0.00	0.23	0.06
R_MW	0.08	0.20	0.02	0.11	0.41	0.10
R_WO	0.00	0.00	0.05	0.00	0.05	0.01
Perimeter	1.38 ± 3.09	1.57 ± 2.23	1.69 ± 3.65	0.69 ± 1.50	5.33 ± 9.24	1.33 ± 2.31
Reference	0.04 ± 0.04	0.10 ± 0.08	0.03 ± 0.03	0.02 ± 0.05	0.19 ± 0.14	0.05 ± 0.04

Table 11. ANCOVA analysis of the effects of site type, conductivity, and their interaction on various ecological measures in reclaimed mine perimeter channels and reference sites. Degrees of freedom = 7. Statistical significance is indicated by *= <0.05, **= <0.005, ***= <0.001.

Parameters	Type	Cond	Type x Cond
Decomp Rate	-0.085	-1.020	3.417*
Mean Total Wt (g) (325 d)	3.394*	4.829**	-1.895
Mean % Organic (325 d)	-2.162	-1.972	5.035**
Mean Organic Mass (g) (325 d)	0.615	1.837	0.744
Mean % Org Mass Lost (325 d)	-0.854	-0.854	4.168**
WVSCI Score	-0.900	-2.673*	5.089**
Pct Chironomid	0.214	1.123	0.030
Pct Tolerant	0.428	1.518	-0.120
Pct EPT	-0.440	-2.420*	2.463*
EPT Richness	-0.877	-3.551**	3.820**
Total Richness	-1.789	0.133	3.318*
Total Inverts	-1.185	-0.520	1.759
Pct 2 Dominant Sp	0.241	1.035	1.056
Total Biomass (g/m^2)	-0.756	-0.935	1.682
Pct Collector- Gatherer	0.175	1.010	0.762
Pct Filterer	-0.465	0.206	0.285
Pct Scraper	-1.636	-1.562	2.009
Pct Shredder	0.593	-0.566	0.601
Pct Predator	0.110	0.606	0.118
Pct Omnivore	-0.989	-0.700	1.180
Pct Unknown	0.915	-0.405	0.416
Amphibian Richness	-1.738	-2.502*	5.533***
Larval Amphibian Richness	0.027*	0.018*	4.775**
Adult Amphibian Richness	0.703	-0.432	1.228
Total No Amph	-1.331	-2.007	3.062*
Mean No Amph	-1.331	-2.007	3.062*
Total No Adult Amph	-0.054	-1.265	1.738
Mean No Adult Amph	0.102	-1.163	1.598
Total No Larval Amph	-2.465*	-2.337	3.511**
Mean No Larval Amph	-1.674	-0.739	-1.469
Total Larval Amph Biomass ($100g/m^2$)	-1.558	-1.058	1.694
Mean Larval Amph Biomass ($100g/m^2$)	-1.558	-1.058	1.694
Mean Amph Density (ind/m^2)	-1.326	-2.003	3.059*
Pct Grassland Amph	-1.018	0.392	0.643
Pct Forest Amph	1.429	1.061	3.041*
Pct Lotic Amph	2.472*	-0.003	0.079
Pct Lentic Amph	-0.723	1.009	1.269

Table 12. Combined larval and adult amphibian density (individuals/m^2) observed on reclaimed mine perimeter channels and reference sites for four sample periods. Mean and standard deviation by site type are given in the last two rows. Perimeter channel sites are listed by increasing age since reclamation.

Site Code	March	May	June	July	Mean Density
P_WO	0.1	0.1	0.1	0.1	0.1
P_AR	0.7	1.3	1.0	1.1	1.0
P_ST	0.0	0.5	0.0	0.0	0.1
P_SU	0.0	0.2	0.3	0.3	0.2
P_BH	0.0	0.6	0.1	0.0	0.2
R_HC	0.0	0.5	0.3	0.0	0.2
R_LF	0.6	0.8	1.2	0.1	0.7
R_ME	1.1	1.2	1.2	0.8	1.0
R_MW	0.1	0.5	0.5	0.5	0.4
R_WO	0.0	0.1	0.1	0.0	0.0
Perimeter	0.2 ± 0.3	0.5 ± 0.5	0.3 ± 0.4	0.3 ± 0.5	0.3 ± 0.4
Reference	0.3 ± 0.5	0.6 ± 0.4	0.6 ± 0.5	0.3 ± 0.3	0.5 ± 0.4

Table 13. Frog and salamander species expected (Exp) to occur in grassland and forest in southwestern West Virginia, based on Green and Pauley (1987) compared to those actually observed (Obs) as (a) adults during visual encounter surveys (VES) (seen or heard), in (l) larval surveys, or (b) for both larval and VES. The preceding "p" indicates individuals encountered in perimeter sites and "r" indicates occurance within reference sites.

		Grassland		Forest	
Aquatic Salamanders		Exp	Obs	Exp	Obs
Appalachian Seal Salamander	*Desmognathus monticola*			x	r.a
Eastern Hellbender	*Cryptobranchus alleganiensis*			x	
Midland Mud Salamander	*Pseudotriton montanus*			x	
Mudpuppy	*Necturus maculosus*	x		x	
Northern Dusky Salamander	*Desmognathus fuscus*			x	r.b
Northern Red Salamander	*Pseudotriton ruber*	x		x	
Northern Two-lined Salamander	*Eurycea bislineata*			x	r.a
Red-spotted Newt	*Notophthalmus v. viridenscens*	x	p.b	x	
Southern Two-lined Salamander	*Eurycea cirrigera*			x	r.l
Spring Salamander	*Gyrinophilus porphyriticus*			x	r.a
Terrestrial Salamanders					
Cumberland Plateau Salamander	*Plethodon kentucki*			x	
Four-toed Salamander	*Hemidactylium scutatum*			x	
Green Salamander	*Aneides aeneus*			x	
Jefferson Salamander	*Ambystoma jeffersonianum*			x	
Longtail Salamander	*Eurycea longicauda*	x		x	
Marbled Salamander	*Ambystoma opacum*			x	
Ravine Salamander	*Plethodon richmondi*			x	
Redback Salamander	*Plethodon cinereus*			x	
Slimy Salamander	*Plethodon glutinosus*			x	
Spotted Salamander	*Ambystoma maculatum*			x	
Wehrle's Salamander	*Plethodon wherlei*			x	
Ambystoma species	*Ambystoma sp.*		p.l	x	
Aquatic Frogs					
Bullfrog	*Rana catesbeiana*	x	p.a	x	
Greenfrog	*Rana clamitans*	x	p.b	x	
Pickerel frog	*Rana palustris*	x	p.a	x	
Northern Leopard Frog	*Rana pipiens*	x		x	
Terrestrial Frogs					
Eastern American Toad	*Bufo americana*	*x*	p.l		
Eastern Spadefoot	*Scaphiopus holbrookii*			x	
Fowler's Toad	*Bufo woodhouseii*				
Gray Treefrog	*Hyla chrysoscelis*		p.l	x	
Mountain Chorus Frog	*Pseudacris brachyphona*			x	
Northern Peeper	*Pseudacris c. cricifer*		p.l	x	r.a
Wood Frog	*Rana sylvatica*			x	

Table 14. Macroinvertebrate measurements from reclaimed mine perimeter channels and reference sites. Mean and standard deviation by site type are given in the last two rows. Perimeter channel sites are listed in increasing age since reclamation. Statistical significance is indicated by *= <0.05.

Site Code	WVSCI Rating	WVSCI Score*	Pct Chironomid	Pct Tolerant	Pct EPT*	EPT Richness*	Total Richness*	Total Inverts	Pct 2 Dominant	# 1 Dominant	# 2 Dominant
P_WO	Poor	33	94	100	0	0	2	1329	98	Chironomidae	Unknown Diptera
P_AR	Marginal	64	10	40	19	1	8	86	39	Snails	Baetidae
P_ST	Marginal	56	29	29	8	1	7	191	75	Cyclopoida	Chironomidae
P_SU	Poor	44	85	87	0	2	10	1384	92	Chironomidae	Cyclopoida
P_BH	Poor	44	72	92	0	0	13	823	86	Chironomidae	Oligochaeta
R_HC	Poor	52	38	58	6	1	6	47	62	Chironomidae	Cyclopoida
R_LF	Excellent	88	0	11	89	6	7	145	57	Ameletidae	Peltoperlidae
R_ME	Good	80	20	23	74	4	7	301	69	Peltoperlidae	Chironomidae
R_MW	Good	76	34	38	52	7	10	512	57	Chironomidae	Ameletidae
R_WO	Poor	45	68	82	18	2	4	60	87	Chironomidae	Capniidae/Leuctridae
Perimeter	-	48 ± 12	58 ± 37	70 ± 33	5 ± 8	1 ± 1	8 ± 4	763 ± 611	78 ± 23	-	-
Reference	-	68 ± 19	32 ± 25	42 ± 28	48 ± 35	4 ± 3	7 ± 2	213 ± 195	66 ± 12	-	-

Table 15. Percent of macroinvertebrates by feeding guild observed on reclaimed mine perimeter channels and reference sites. Guilds include collector gatherer (CG), filterer (FI), scraper (SC), shredder (SH), predator (PR), omnivore (OM), and unknown (UN). Mean and standard deviation by site type are given in the last two rows. Perimeter channel sites are listed in increasing age since reclamation. Statistical significance is indicated by *= <0.05.

Site Code	CG	FI	SC	SH	PR	OM	UN*
P_WO	94	0	0	0	6	0	0
P_AR	30	0	21	9	21	19	0
P_ST	70	2	2	0	4	22	0
P_SU	93	0	6	0	1	0	1
P_BH	82	8	2	0	8	0	0
R_HC	55	0	17	6	2	17	2
R_LF	61	0	0	20	14	0	6
R_ME	25	0	0	69	0	0	5
R_MW	61	0	0	22	6	0	10
R_WO	82	0	0	18	0	0	0
Perimeter	74 ± 26	2 ± 3	6 ± 9	2 ± 4	8 ± 8	8 ± 11	0 ± 0
Reference	57 ± 20	0 ± 0	3 ± 8	27 ± 24	5 ± 6	3 ± 8	5 ± 4

Table 16. Mean organic matter transport distances and retention rate for reclaimed mine perimeter channels and reference sites. Mean and standard deviation by site type are given in the last two rows. Perimeter channel sites are listed in increasing age since reclamation. Statistical significance is indicated by *= <0.05.

Site Code	Mean Leaf Distance (m)	Mean Cum Stick Distance (m)*	Mean Cum Stick Dist/Day*	Retention Rate	Pct Sticks Retained*	Dowels Exiting Reach*	Gradient
P_WO	2.6	37.2	0.2	-0.0089	36	32	6
P_AR	0.0	0.0	0.0	-0.0782	100	0	1
P_ST	0.0	0.0	0.0	-0.0782	100	0	1
P_SU	0.0	0.0	0.0	-0.0782	100	0	1
P_BH	0.0	0.0	0.0	-0.0782	100	0	1
R_HC	0.4	11.7	0.1	-0.0367	84	8	17
R_LF	0.2	30.2	0.2	-0.0204	64	18	6
R_ME	0.0	22.9	0.1	-0.0216	66	17	16
R_MW	0.8	28.6	0.1	-0.0147	52	24	8
R_WO	0.8	41.1	0.2	-0.0066	28	36	7
Perimeter	0.5 ± 1.2	7.4 ± 16.6	0.0 ± 0.1	-0.0644 ± 0.0310	87 ± 29	6 ± 14	2 ± 2
Reference	0.4 ± 0.3	26.9 ± 10.7	0.1 ± 0.1	-0.0200 ± 0.0111	59 ± 21	21 ± 10	11 ± 5

Table 17. Dissolved organic carbon measures (mg/L) for reference sites and reclaimed mine perimeter channels. Reference sites did not contain water at the time of autumn sampling. Autumn samples for P_ST were contaminated. Site mean is the mean of spring, summer, and winter only. Mean and standard deviation by site type are given in the last two rows. Perimeter channel sites are listed in increasing age since reclamation. Statistical significance is indicated by *= <0.05.

Site Code	Spring	Summer	Autumn	Winter*	Site Mean
P_WO	0.42	1.10	1.29	1.18	0.90
P_AR	5.71	1.53	2.99	3.20	3.48
P_ST	7.74	2.34	-	2.94	4.34
P_SU	8.99	7.49	8.01	1.72	6.07
P_BH	1.72	1.47	4.41	1.52	1.57
R_HC	0.32	0.27	-	0.67	0.42
R_LF	3.43	1.42	-	0.98	1.94
R_ME	2.61	0.86	-	0.92	1.46
R_MW	2.77	1.39	-	1.18	1.78
R_WO	3.37	1.29	-	1.12	1.93
Perimeter	4.92 ± 3.73	2.79 ± 2.65	4.18 ± 3.10	2.11 ± 0.90	3.27 ± 2.09
Reference	2.50 ± 1.27	1.05 ± 0.49	-	0.97 ± 0.20	1.51 ± 0.64

Table 18. Total dissolved carbon measures (mg/L) for reference sites and reclaimed mine perimeter channels. Reference sites did not contain water at the time of autumn sampling. Autumn samples for P_ST were contaminated. Site mean is the mean of spring, summer, and winter only. Mean and standard deviation by site type are given in the last two rows. Perimeter channel sites are listed in increasing age since reclamation. Statistical significance is indicated by *= <0.05.

Site Code	Spring*	Summer*	Autumn	Winter*	Site Mean*
P_WO	44.4	26.3	2.0	2.1	24.3
P_AR	28.1	18.9	2.9	2.9	16.6
P_ST	26.1	33.7	-	3.0	20.9
P_SU	22.8	29.0	5.1	5.8	19.2
P_BH	13.6	18.3	3.3	3.1	11.7
R_HC	1.2	2.1	-	1.1	1.5
R_LF	5.3	3.2	-	1.4	3.3
R_ME	4.0	3.7	-	1.5	3.0
R_MW	4.1	3.1	-	1.6	2.9
R_WO	4.1	1.7	-	1.6	2.5
Perimeter	27.0 ± 5.0	25.2 ± 3.0	3.3 ± 0.6	3.4 ± 0.6	18.5 ± 2.1
Reference	3.7 ± 0.7	2.7 ± 0.4	-	1.4 ± 0.1	2.6 ± 0.3

Table 19. Mean total weight (g), mean organic (g) and inorganic mass (g), percent organic, percent organic mass lost, decomposition rate (k). and processing power observed on reclaimed mine perimeter channels and reference sites after ~325 days of exposure. Mean and standard deviation by site type are given in the last two rows. Perimeter channel sites are listed in increasing age since reclamation. Statistical significance is indicated by *= <0.05.

Site Code	Days of Exposure	Mean Total Weight*	Mean Organic Mass	Mean Inorganic Mass	Mean % Organic*	Mean % Organic Mass Lost*	Decomp Rate*	Process Power *100
P_WO	199	10.2	5.6	4.6	57	45	-0.00268	0.002
P_AR	325	5.8	5.1	0.7	88	50	-0.00231	0.018
P_ST	328	6.0	5.5	0.5	92	45	-0.00167	0.013
P_SU	329	5.6	5.2	0.4	92	49	-0.00205	0.016
P_BH	325	5.7	5.4	0.3	94	47	-0.00194	0.015
R_HC	200	11.0	6.0	5.0	57	41	-0.00248	0.009
R_LF	327	1.4	1.1	0.3	82	89	-0.00666	0.014
R_ME	329	3.4	2.9	0.5	85	71	-0.00380	0.008
R_MW	328	5.1	3.8	1.4	75	63	-0.00297	0.004
R_WO	326	5.6	5.3	0.3	94	48	-0.00149	0.001
Perimeter		6.7 ± 2.0	5.3 ± 0.2	1.3 ± 1.8	85 ± 16	47 ± 2	-0.00213 ± 0.00038	0.013 ± 0.006
Reference		5.3 ± 3.6	3.8 ± 1.9	1.5 ± 2.0	78 ± 14	62 ± 19	-0.00348 ± 0.00196	0.007 ± 0.005

Table 20. Correlation of parameters with decomposition rates and mean conductivity for reference sites and reclaimed mine perimeter channels. Mean and standard error are given in the first two rows.

Response Variables	Perimeter Mean	Reference Mean	Correlation with Decomp	Correlation with Conductivity
Mean Q (m^3/s)	0.009 (0.003)	0.002 (0.001)		0.76
Water Chemistry				
Mean Alkalinity (mg/L)	138 (13)	5 (1)		0.84
Mean Ca (mg/L)	163 (31)	21 (15)		0.99
Mean Mg (mg/L)	154 (34)	9 (4)		0.96
Mean SO_4 (mg/L)	1008 (196)	32 (20)		0.89
Mean Summer pH	7.2 (0.2)	5.7 (0.3)		0.80
Mean Spr Cond (µS/cm)	2310 (495)	602 (445)		0.95
Mean Sum Cond (µS/cm)	2255 (488)	642 (501)		0.93
Mean Aut Cond (µS/cm)	2147 (448)	133 (75)		0.89
Mean Win Cond (µS/cm)	2077 (395)	147 (64)		0.93
Mean Cond (µS/cm)	2197 (414)	461 (326)		1.00
Spring TDS (mg/L)	1366 (294)	76 (28)		0.91
Summer TDS (mg/L)	1818 (455)	157 (95)		0.96
Autumn TDS (mg/L)	-	-		-
Winter TDS (mg/L)	1317 (269)	77 (33)		0.91
Mean TDS (mg/L)	1501 (279)	103 (50)		0.97
Spring TC (mg/L)	27.0 (5.0)	3.7 (0.7)		0.78
Summer TC (mg/L)	25.2 (3.0)	2.7 (0.4)		0.80
Mean TC (mg/L)	18.5 (2.1)	2.6 (0.3)		0.81
Habitat Assessment				
EPA RBP	78 (6)	150 (10)		-0.78
WV FCU	3 (0)	9 (0)		-0.84
ORAM	35 (6)	61 (4)		-0.80

Table 20 continuted.

Response Variables	Perimeter Mean	Reference Mean	Correlation with Decomp	Correlation with Conductivity
Vegetation				
Pct Open Water	13 (6)	0 (0)		0.82
Species per km^2	0.1 (0.1)	2.5 (0.3)		-0.80
Pct Canopy Cover	4 (4)	91 (1)		-0.82
Macroinvertebrates				
WVSCI Score	48 (5)	68 (8)	0.75	
Pct EPT	5 (4)	48 (16)	0.86	-0.75
EPT Richness	1 (0)	4 (1)		-0.80
Pct Predator	8 (3)	5 (3)		0.95
Pct Unknown	0 (0)	5 (2)	0.83	
Amphibians				
Total Adult Amph Sp	1 (0)	2 (0)	0.78	
Total No Adult Amph	5 (3)	28 (13)	0.80	
Pct Lotic Amph	5 (3)	86 (13)		-0.75
Pct Lentic Amph	89 (8)	54 (8)		0.75
OM Processing				
Decomp Rate	0.0021 (0.0002)	0.0035 (0.0009)	1.00	
Mean Total Wt (g) (325 d)	6.7 (0.9)	5.3 (1.6)		0.77
Mean Organic Mass (g) (325 d)	5.3 (0.1)	3.8 (0.9)	-0.94	
Mean % Org Mass Lost (325 d)	47 (1)	62 (9)	0.95	
OM Retention				
Mean Cum Stick Distance (m)	7.44 (7.44)	26.89 (4.81)	0.76	
Mean Cum Stick Dist/ Day	0.04 (0.04)	0.14 (0.02)	0.76	

Table 21. Ecological units (EU) ratios, EU_I ratios (calculated using ideal reference means), perimeter means, reference means, and ideal reference means for reclaimed mine perimeter channels and reference sites. Species parameters are standardized by the area of aquatic feature sampled.

Response Variables	Perimeter Mean	Reference Mean	Ideal Ref Mean	EU Ratio	EU_I Ratios
Mean Larval Amph Biomass (100g/m^2)	1.33 (1.03)	0.05 (0.02)	0.07 (0.02)	28	19
Mean Cond (µS/cm)	2197 (414)	461 (326)	61 (2)	5	36
Retention Rate	-0.0644 (0.0139)	-0.0199 (0.0049)	-0.0189 (0.0021)	3.2	3.4
Mean DOC (mg/L)	3.51 (0.94)	1.51 (0.28)	1.73 (0.14)	2.3	2.0
Processing Power *100	0.013 (0.003)	0.007 (0.002)	0.009 (0.003)	1.8	1.5
Pct Lentic Amph	89 (8)	54 (8)	61 (11)	0.97	0.86
Total Invert Biomass (g/m^2)	31.8 (18.9)	34.5 (14.9)	44.2 (22.3)	0.92	0.72
WVSCI Score	48 (5)	68 (8)	81 (3)	0.71	0.59
Total Invertebrate Richness	8 (2)	7 (1)	8 (1)	0.71	0.60
Decomp Rate	0.0021 (0.0002)	0.0035 (0.0009)	0.0045 (0.0011)	0.61	0.48
EPA RBP	78 (6)	150 (10)	134 (1)	0.52	0.59
EPT Richness	1 (0)	4 (1)	6 (1)	0.13	0.09
Pct EPT	5 (4)	48 (16)	72 (11)	0.10	0.06
Pct Lotic Amph	5 (3)	86 (13)	99 (1)	0.03	0.03

Figures

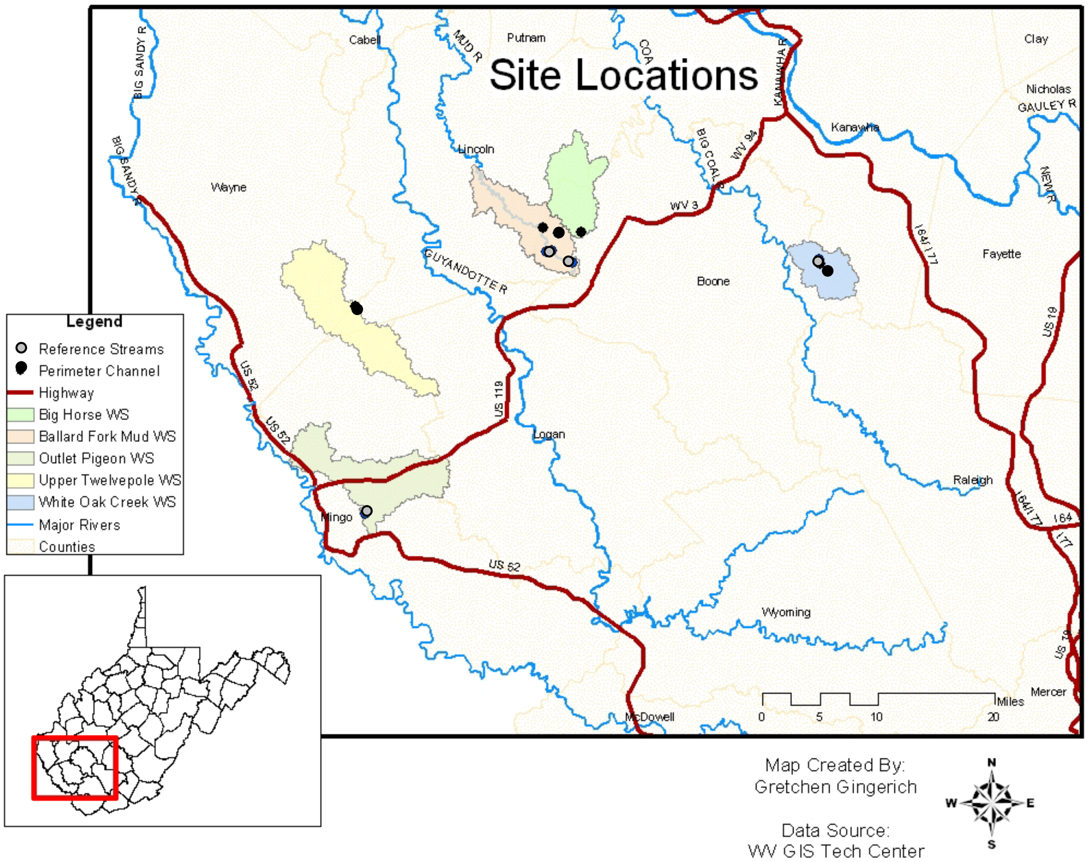

Figure 1. Site locations and HUC 12 watersheds for reference sites (gray dots) and perimeter channels (black dots).

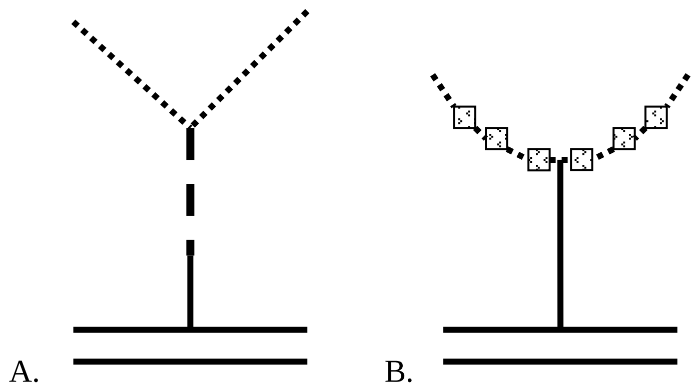

Figure 2. A.) A typical headwater system: two ephemeral streams (dotted line) feeding into an intermittent (dashed line), feeding into a perennial (solid line), and finally to a broad river. B.) A series of sediment ponds, or wetlands, on the perimeter of a valley-fill system. This system usually has an intermittent or perennial outflow off-site to a larger system.

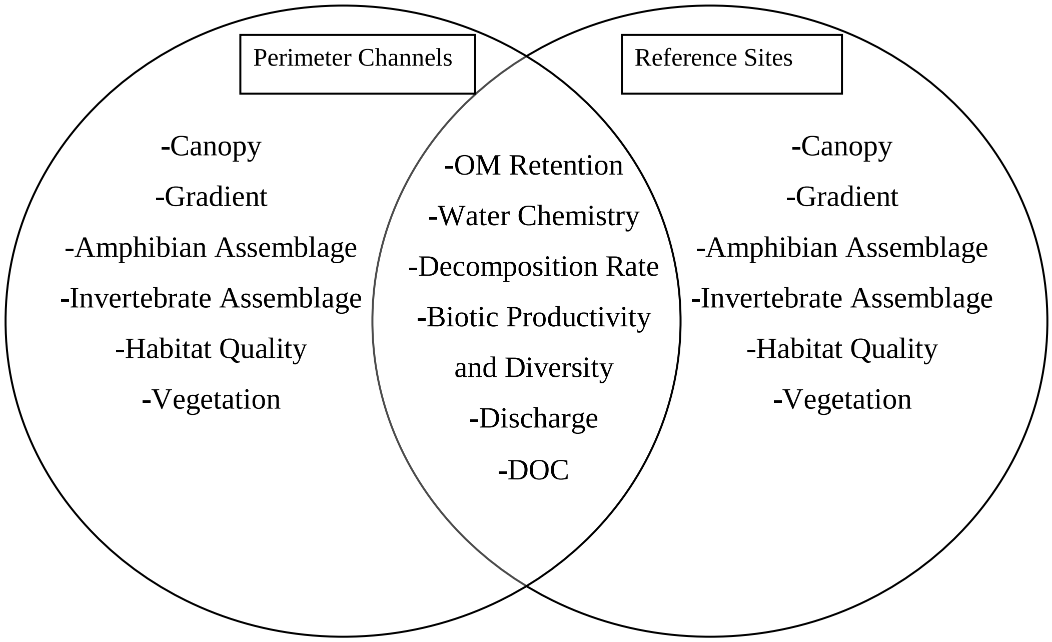

Figure 3. Directly comparable and indirectly comparable site parameters for reference and reclaimed surface mine sites.

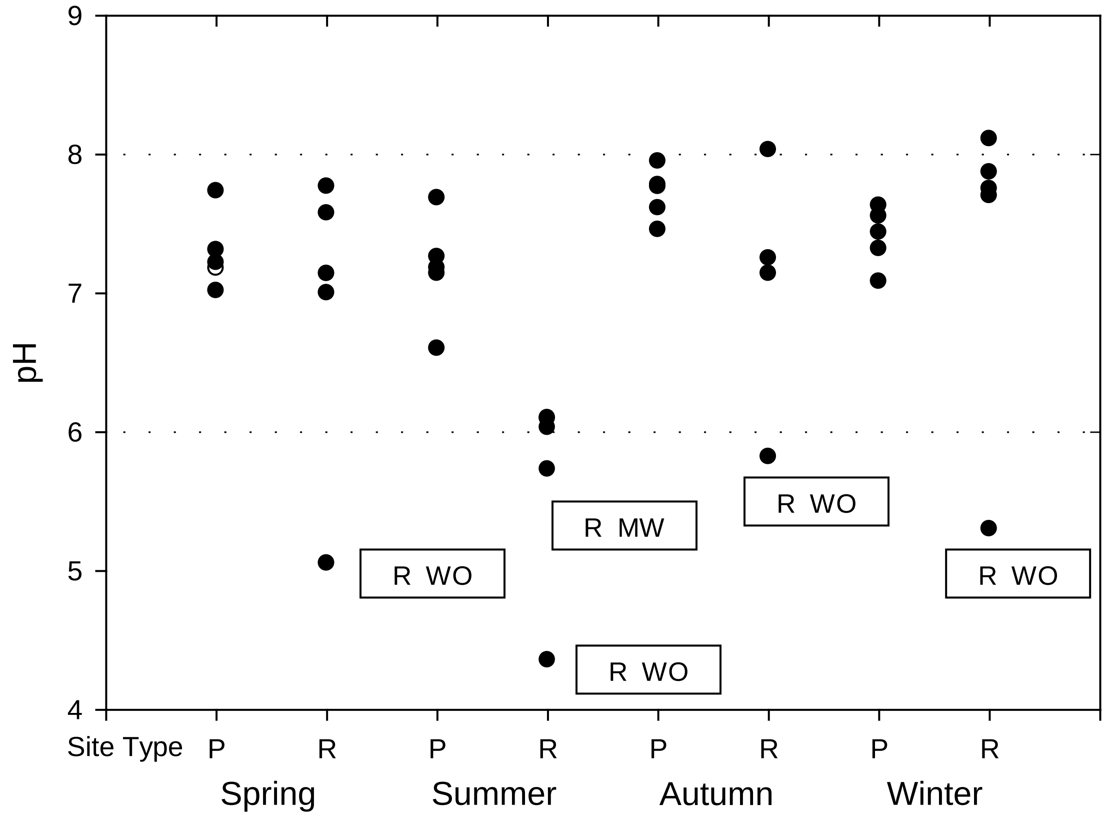

Figure 4. Seasonal pH for reclaimed mine perimeter channels and reference streams combined by site type. A range of pH 6.0-8.0 (dashed lines) is considered normal or acceptable.

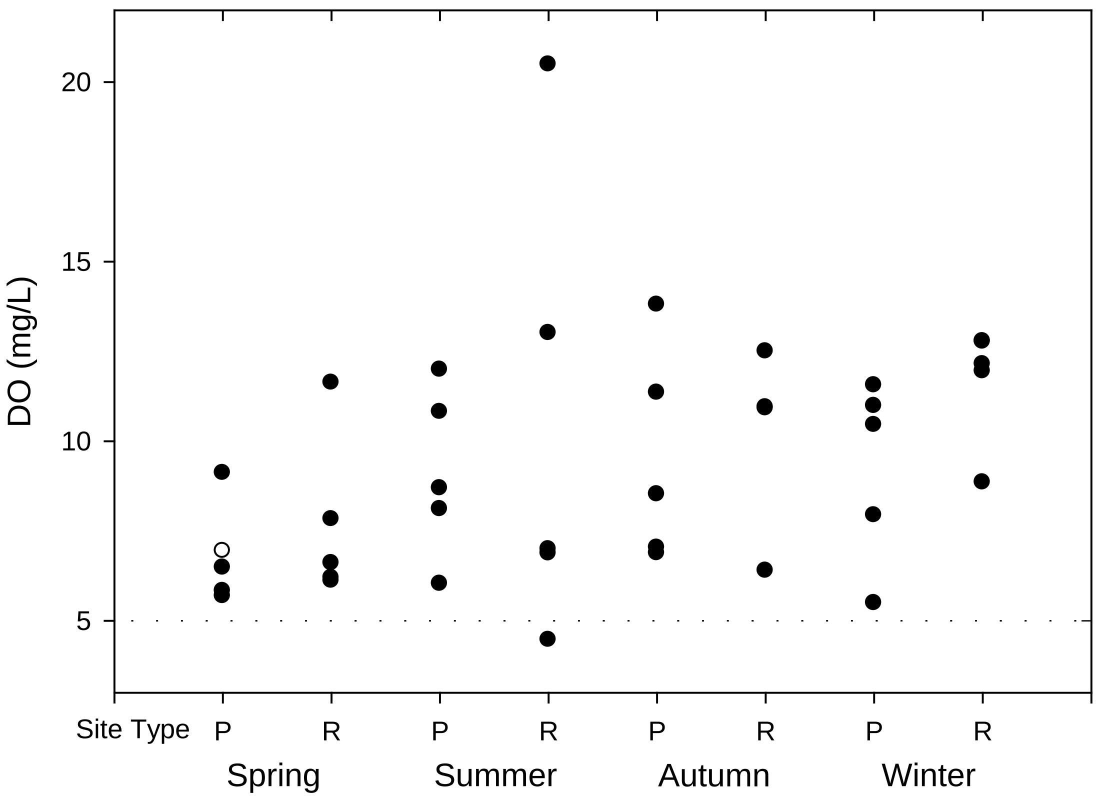

Figure 5. Seasonal dissolved oxygen (mg/L) for reclaimed mine perimeter channels and reference streams combined by site type. A limit of 5.0 mg/L (dashed line) is recommended for the health of aquatic life.

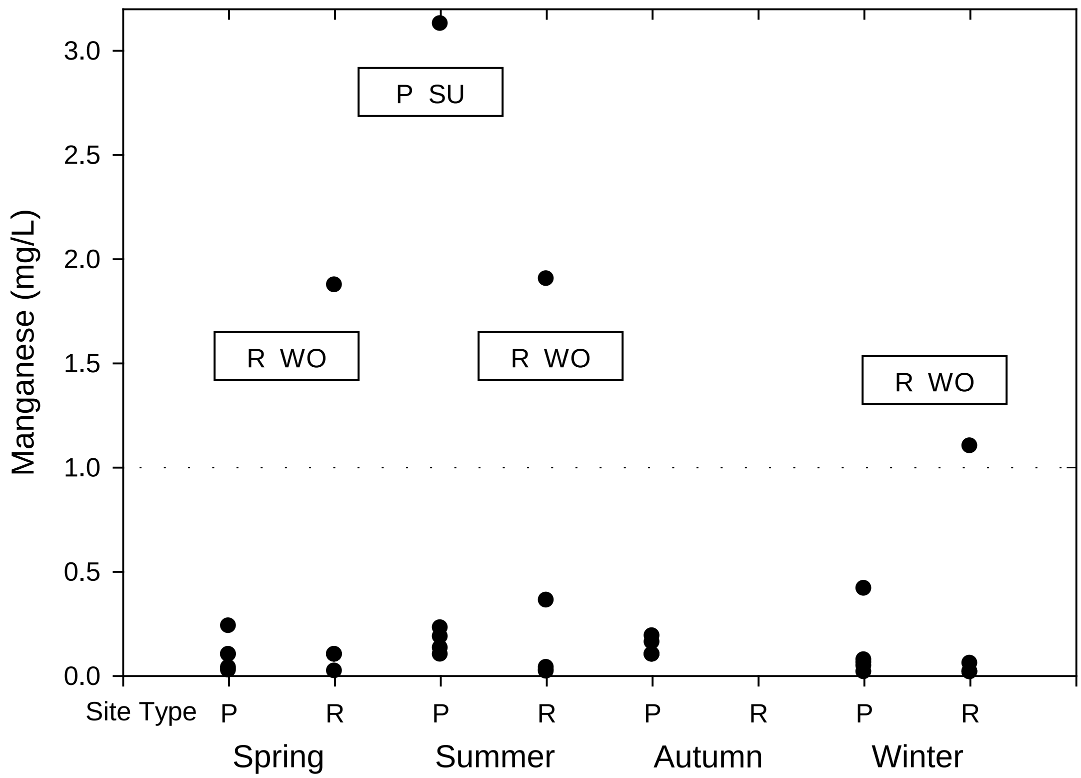

Figure 6. Seasonal manganese (mg/L) for reclaimed mine perimeter channels and reference streams combined by site type. The WWF limit is 1 mg/L (dashed line). Method detection limits (MDL) were 0.017 mg/L. Reference sites did not contain enough water for sampling in autumn.

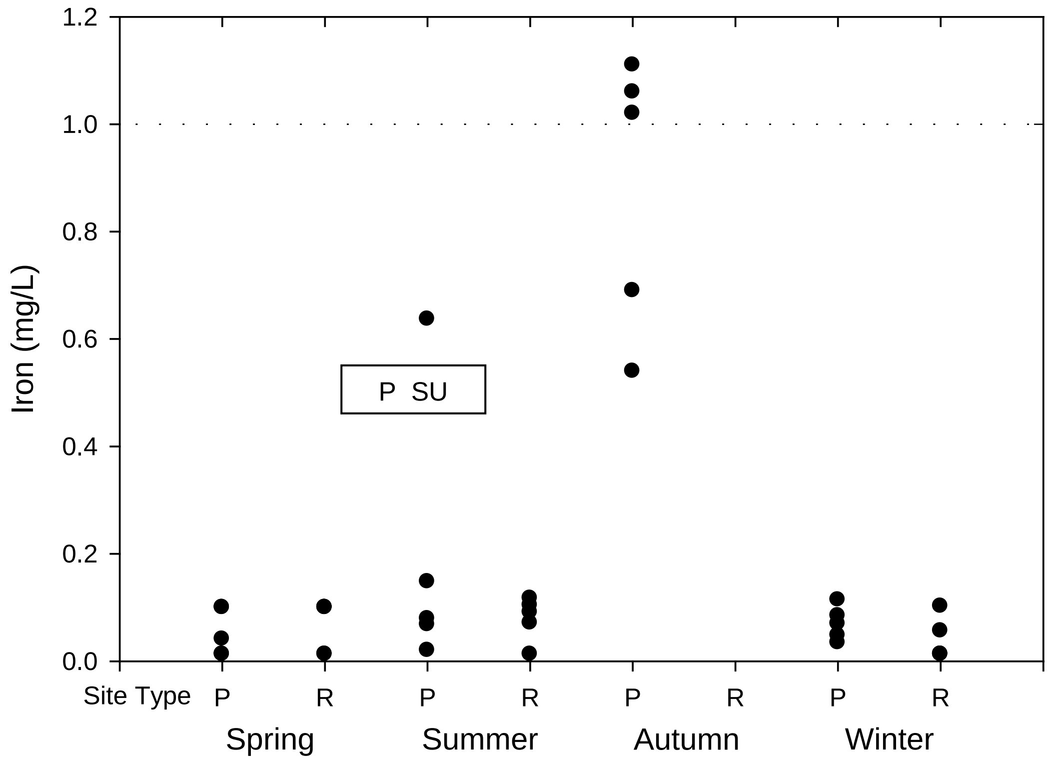

Figure 7. Seasonal iron (mg/L) for reclaimed mine perimeter channels and reference streams combined by site type. A limit of 1.0 mg/L is recommended for the health of aquatic life (dashed line). Reference sites did not contain enough water for sampling in autumn.

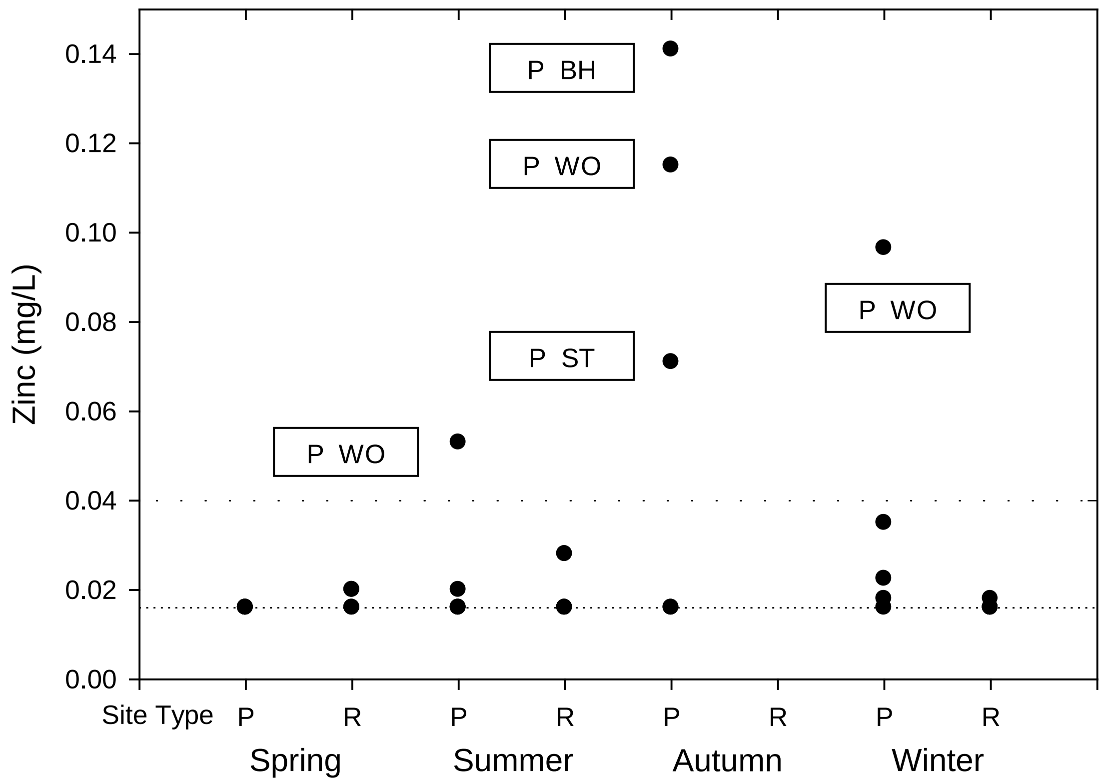

Figure 8. Seasonal zinc (mg/L) for reclaimed mine perimeter channels and reference streams combined by site type. MDL was 0.016 mg/L (dotted line). The recommended level for the health of aquatic life is 0.04 mg/L (dashed line). Reference sites did not contain enough water for sampling in autumn.

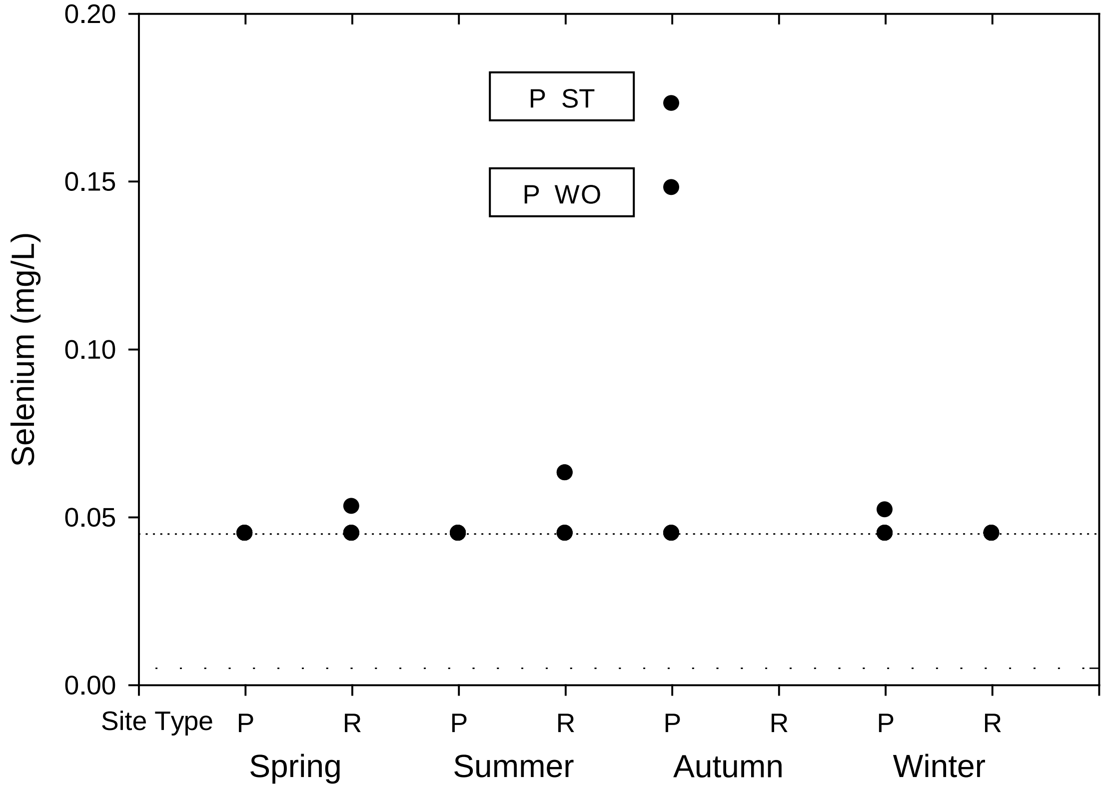

Figure 9. Seasonal selenium (mg/L) for reclaimed mine perimeter channels and reference streams combined by site type. The WWF limit is 0.005 mg/L (dashed line). MDL was 0.045 mg/L (dotted line). Reference sites did not contain enough water for sampling in autumn.

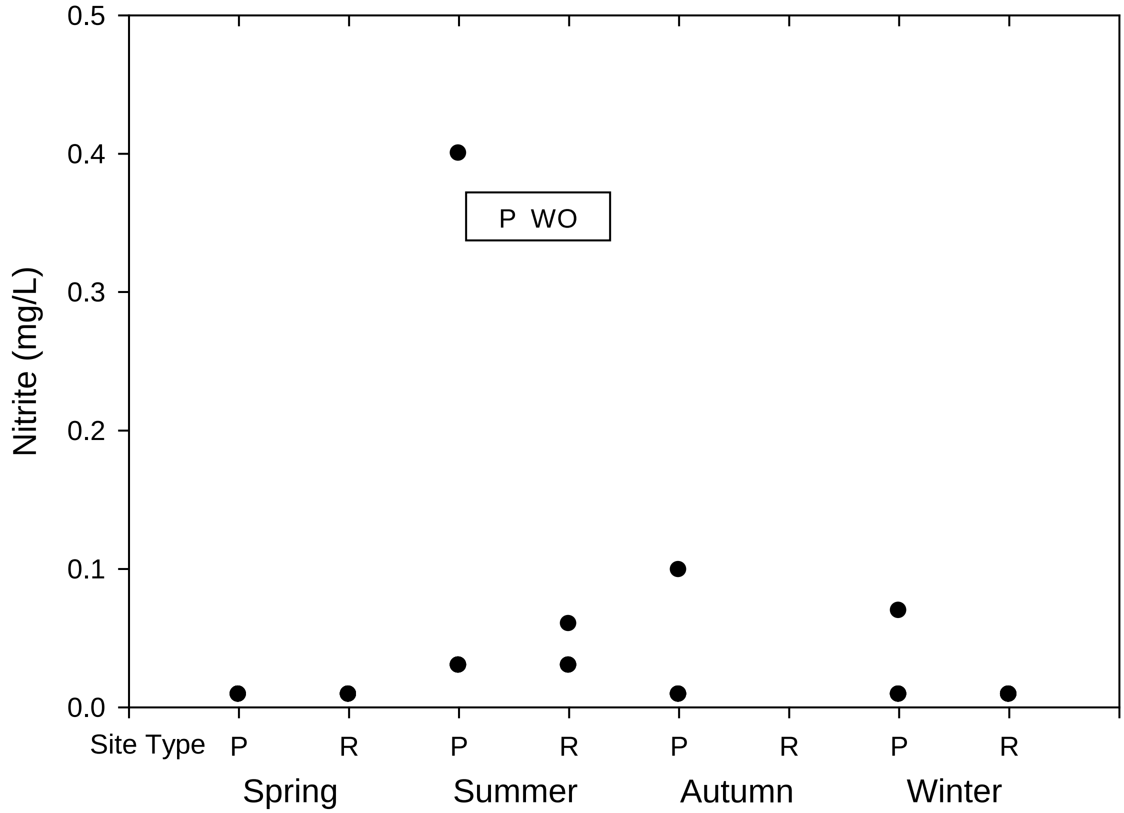

Figure 10. Seasonal nitrite (mg/L) for reclaimed mine perimeter channels and reference streams combined by site type. R_HC experienced a summer measure of 46.230 mg/L (not shown) after disturbance. Reference sites did not contain enough water for sampling in autumn.

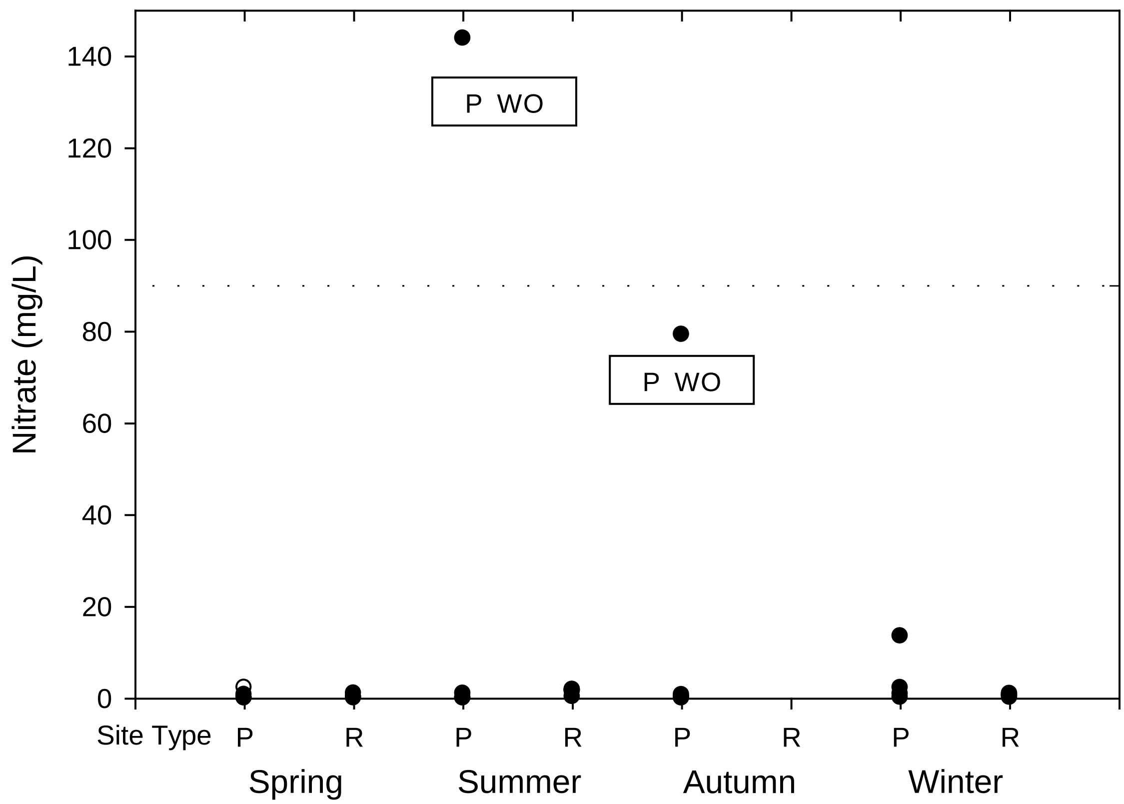

Figure 11. Seasonal nitrate (mg/L) for reclaimed mine perimeter channels and reference streams combined by site type. The WWF limit is 90 mg/L (dashed line). Reference sites did not contain enough water for sampling in autumn.

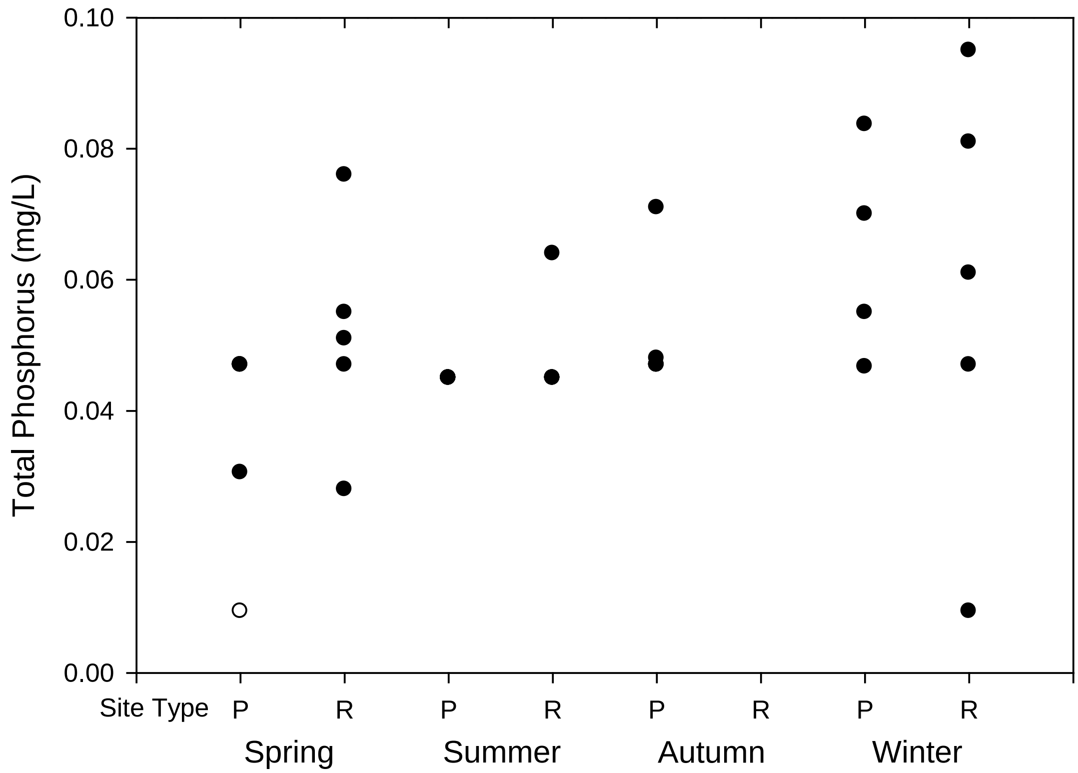

Figure 12. Seasonal total phosphorus (mg/L) for reclaimed mine perimeter channels and reference streams combined by site type. In 1986, EPA recommended a 0.1 mg/L limit for streams not emptying into reservoirs. Reference sites did not contain enough water for sampling in autumn.

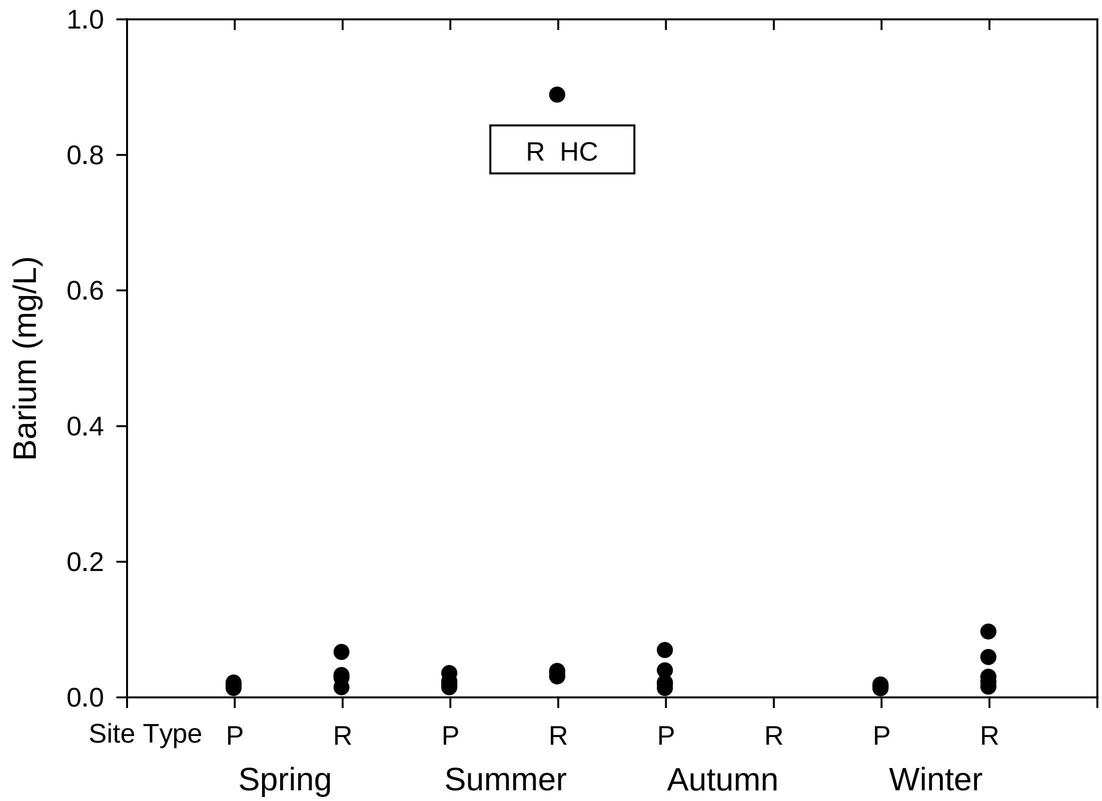

Figure 13. Seasonal barium (mg/L) for reclaimed mine perimeter channels and reference streams combined by site type. Reference sites did not contain enough water for sampling in autumn.

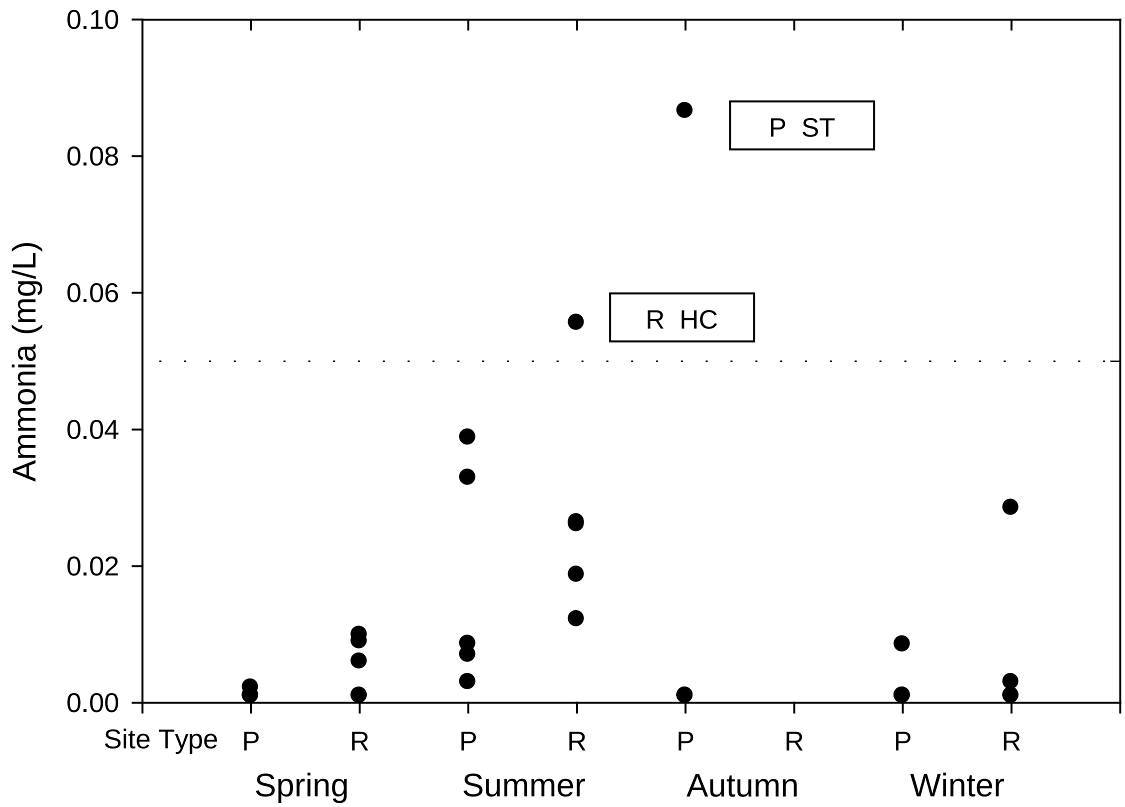

Figure 14. Seasonal ammonia (mg/L) for reclaimed mine perimeter channels and reference streams combined by site type. The recommended limit for aquatic life health is 0.05 mg/L (dashed line). Reference sites did not contain enough water for sampling in autumn.

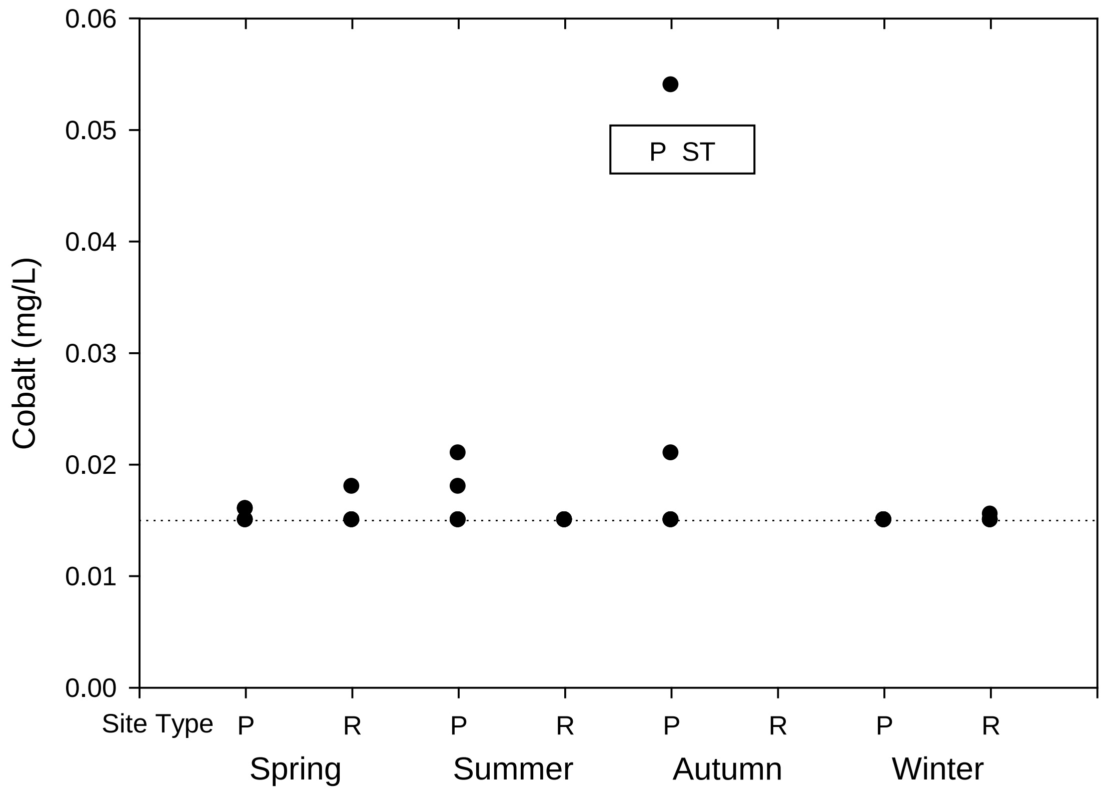

Figure 15. Seasonal cobalt (mg/L) for reclaimed mine perimeter channels and reference streams combined by site type. There is no recommended limit for aquatic health. MDL was 0.015 mg/L (dotted line). Reference sites did not contain enough water for sampling in autumn.

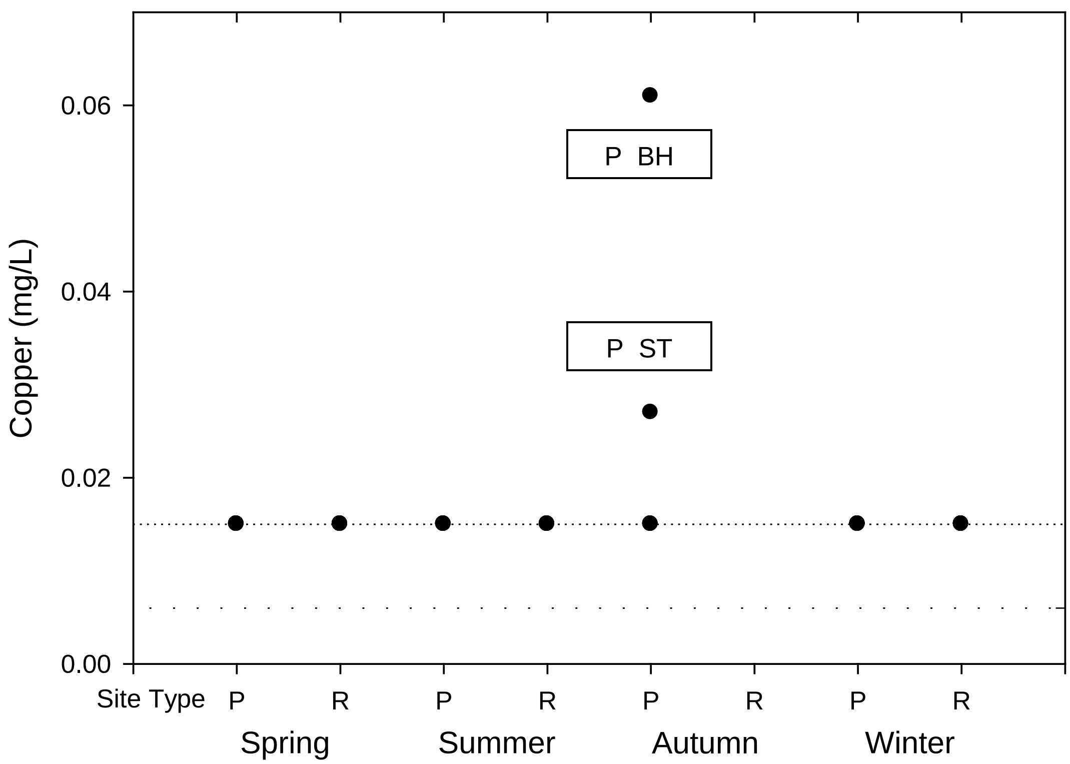

Figure 16. Seasonal copper (mg/L) for reclaimed mine perimeter channels and reference streams combined by site type. The WWF limit is 0.006 mg/L (dashed line). MDL was 0.015 mg/L (dotted line). Reference sites did not contain enough water for sampling in autumn.

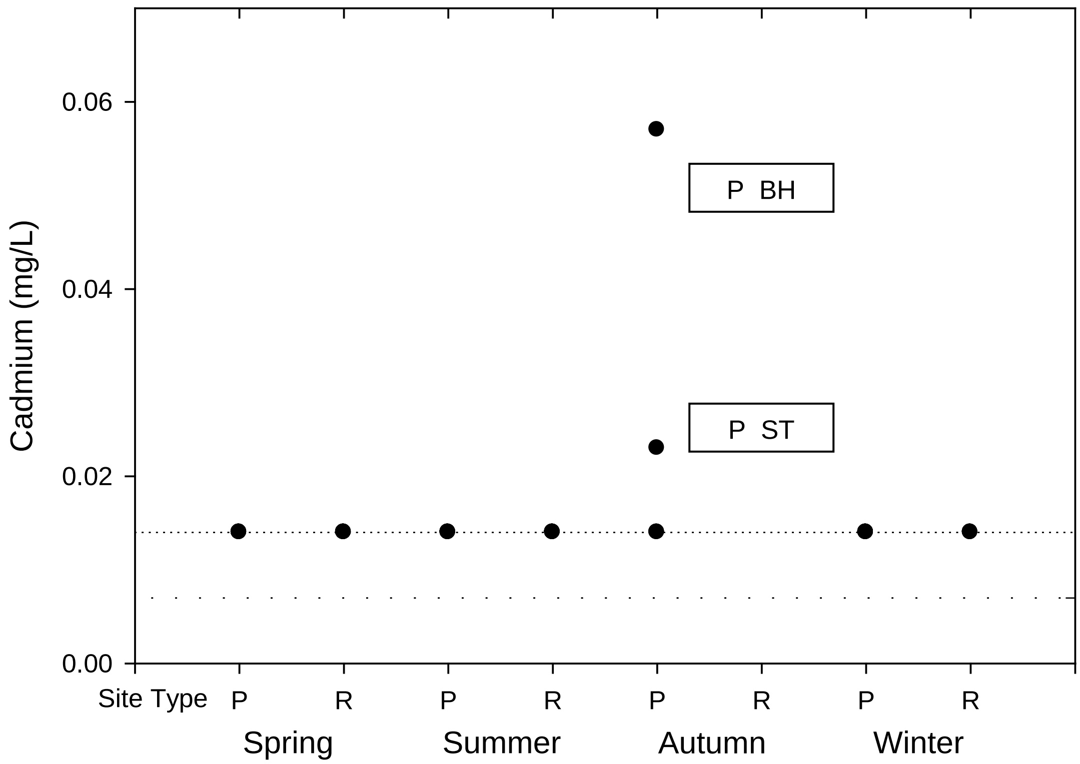

Figure 17. Seasonal cadmium (mg/L) for reclaimed mine perimeter channels and reference streams combined by site type. The WWF limit is 0.007 mg/L (dashed line). However, method detection limit (MDL) was 0.014 (dotted line). Reference sites did not contain enough water for sampling in autumn.

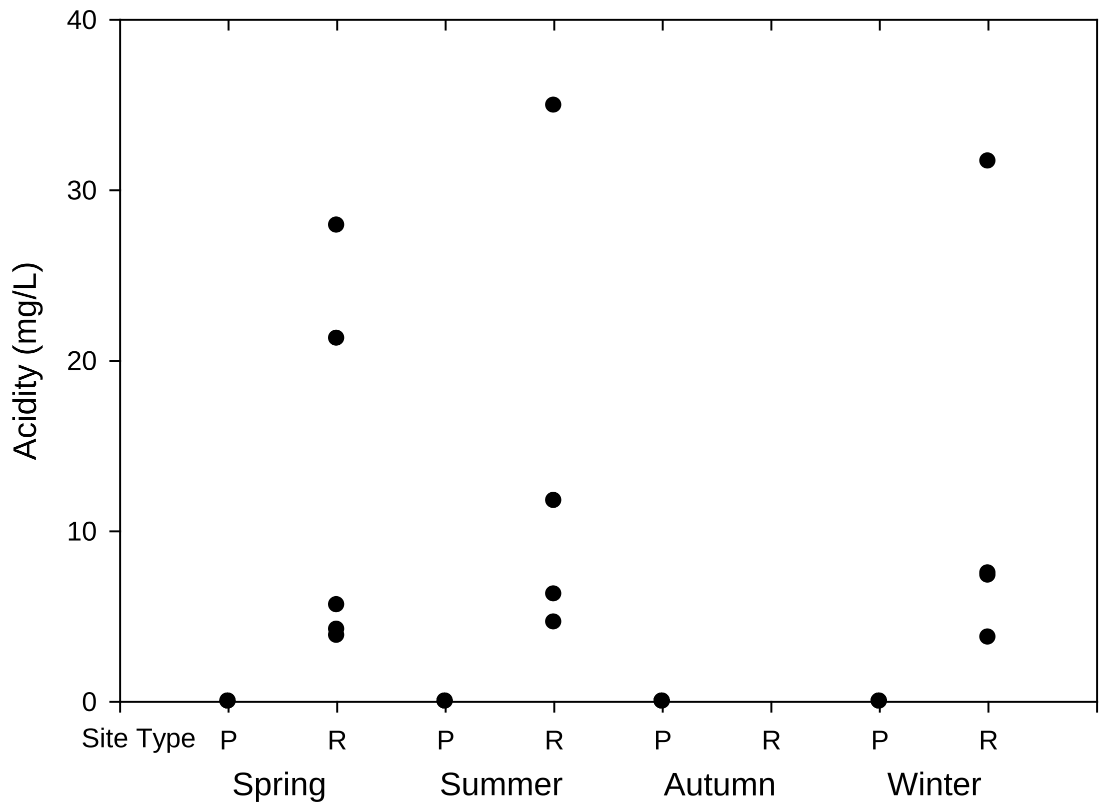

Figure 18. Seasonal acidity (mg/L) for reclaimed mine perimeter channels and reference streams combined by site type. Reference sites did not contain enough water for sampling in autumn.

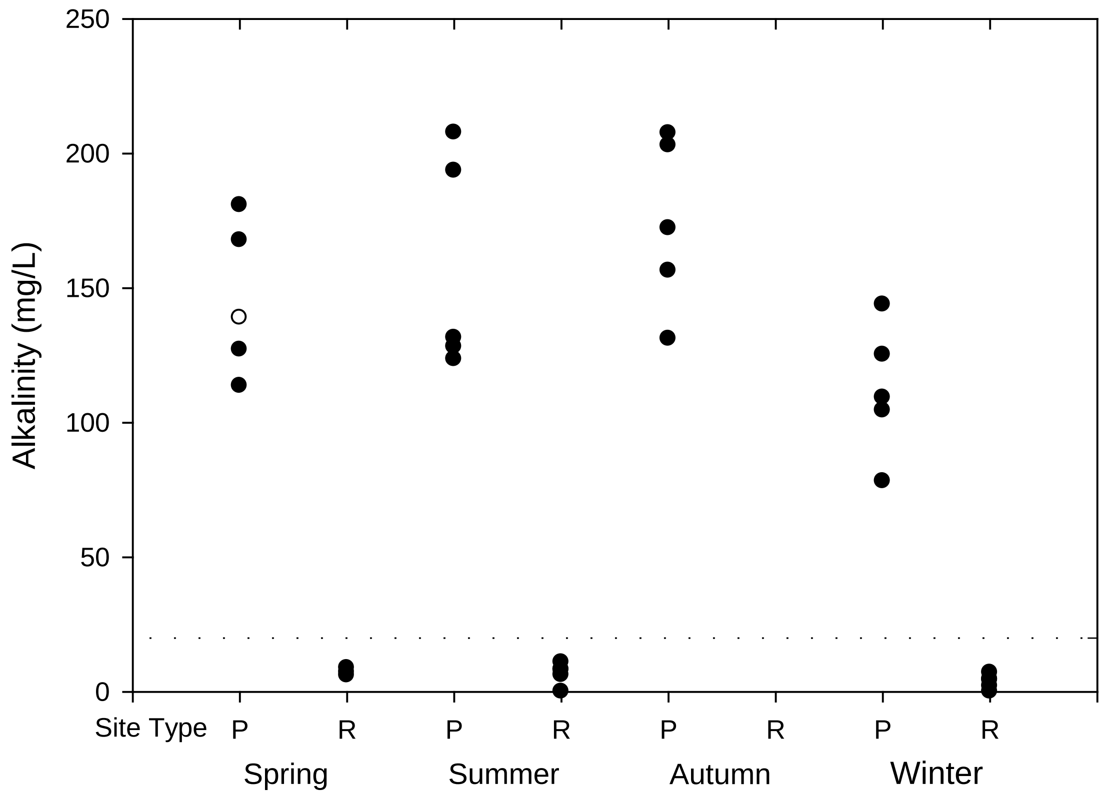

Figure 19. Seasonal alkalinity (mg/L) for reclaimed mine perimeter channels and reference streams combined by site type. An alkalinity of > 20mg/L (dashed line) is considered to have good buffering capacity. Reference sites did not contain enough water for sampling in autumn.

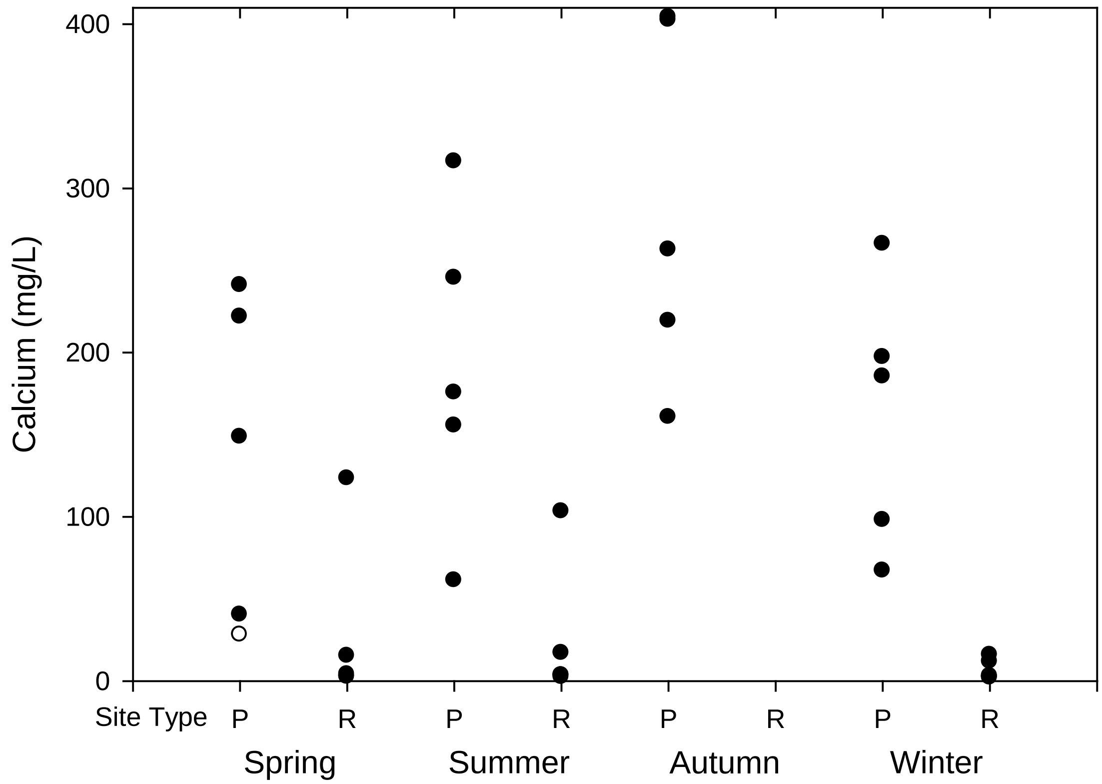

Figure 20. Seasonal calcium (mg/L) for reclaimed mine perimeter channels and reference streams combined by site type. Reference sites did not contain enough water for sampling in autumn.

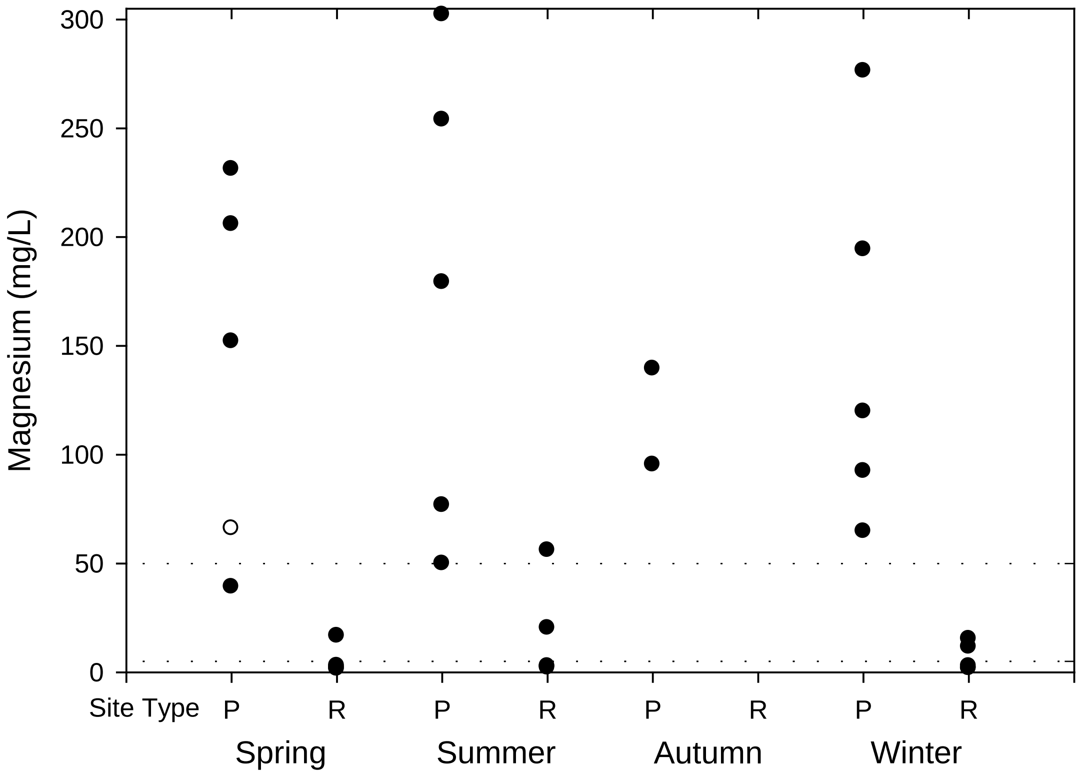

Figure 21. Seasonal magnesium (mg/L) for reclaimed mine perimeter channels and reference streams combined by site type. Streams with magnesium sources usually have levels of 5-50 mg/L (dashed lines). Reference sites did not contain enough water for sampling in autumn.

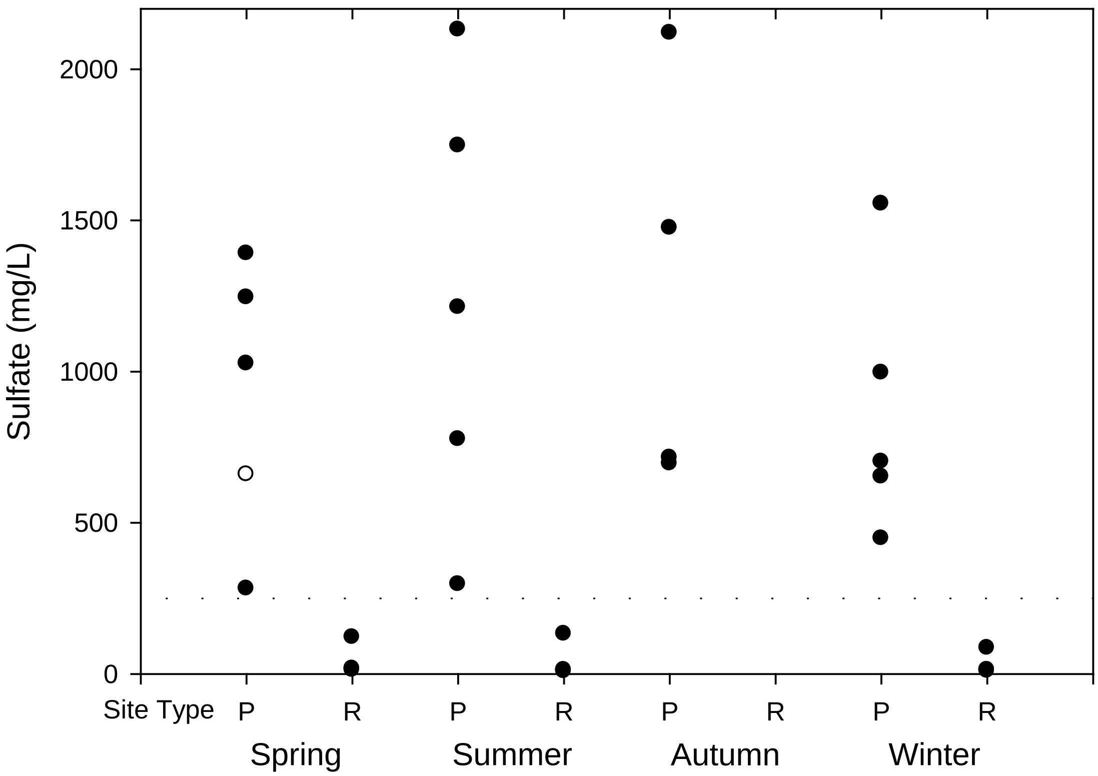

Figure 22. Seasonal sulfate (mg/L) for reclaimed mine perimeter channels and reference streams combined by site type. Perimeter sites measured above 250 mg/L (dashed line). Reference sites did not contain enough water for sampling in autumn.

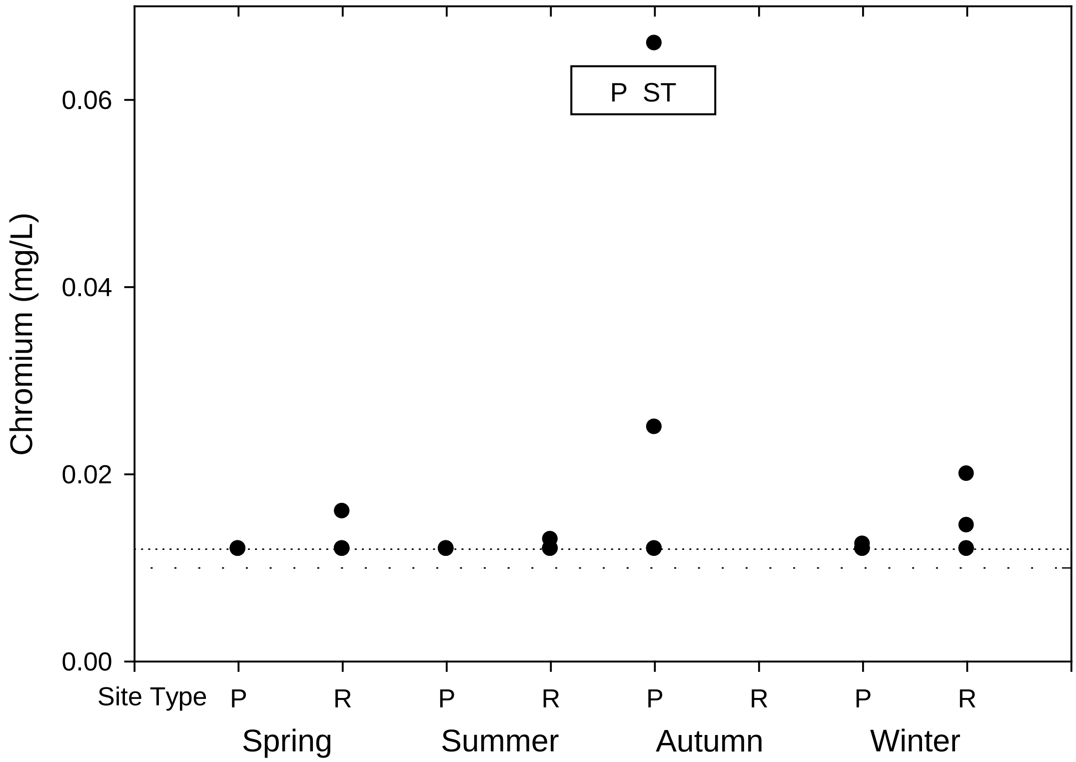

Figure 23. Seasonal chromium (mg/L) for reclaimed mine perimeter channels and reference streams combined by site type. The WWF limit for chromium is 0.01 mg/L (dashed line). MDL was 0.012 mg/L (dotted line). Reference sites did not contain enough water for sampling in autumn.

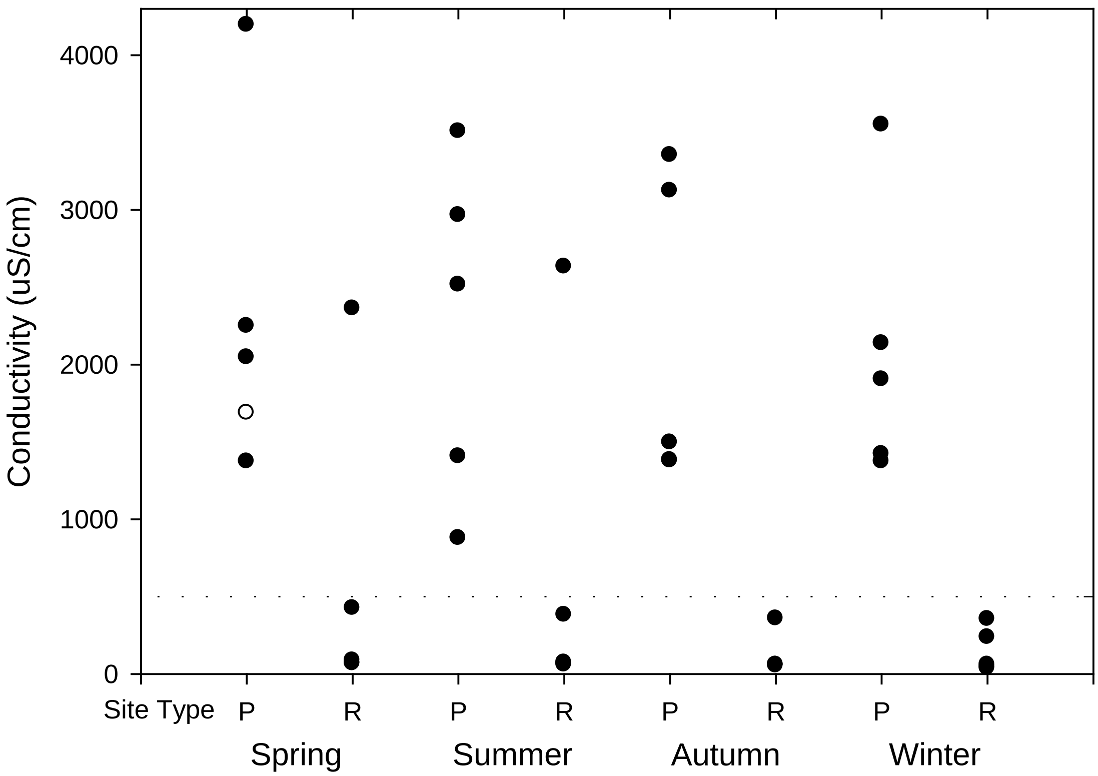

Figure 24. Seasonal conductivity (µS/cm) for reclaimed mine perimeter channels and reference streams combined by site type. A conductivity of 500 µS/cm (dashed line) is the EPA recommended upper limit for healthy fisheries.

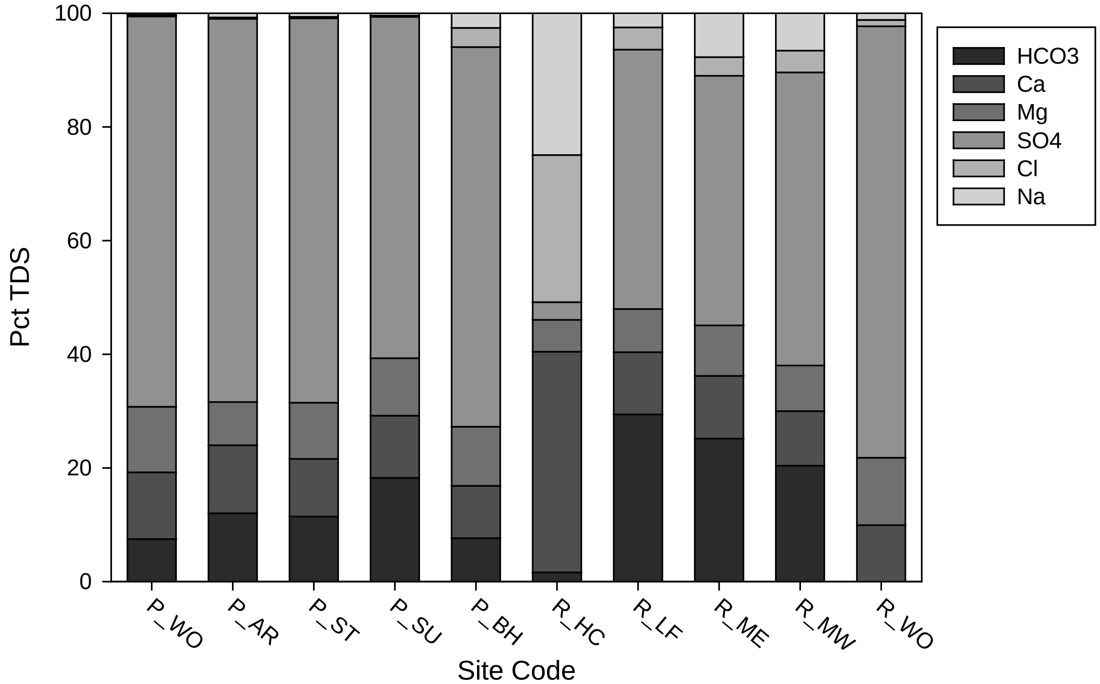

Figure 25. The percent of mean TDS that was composed of bicarbonate, calcium, magnesium, sulfate, chloride, and sodium for reclaimed surface mine perimeter channel and reference sites. Perimeter channels are presented in order of age since reclamation.

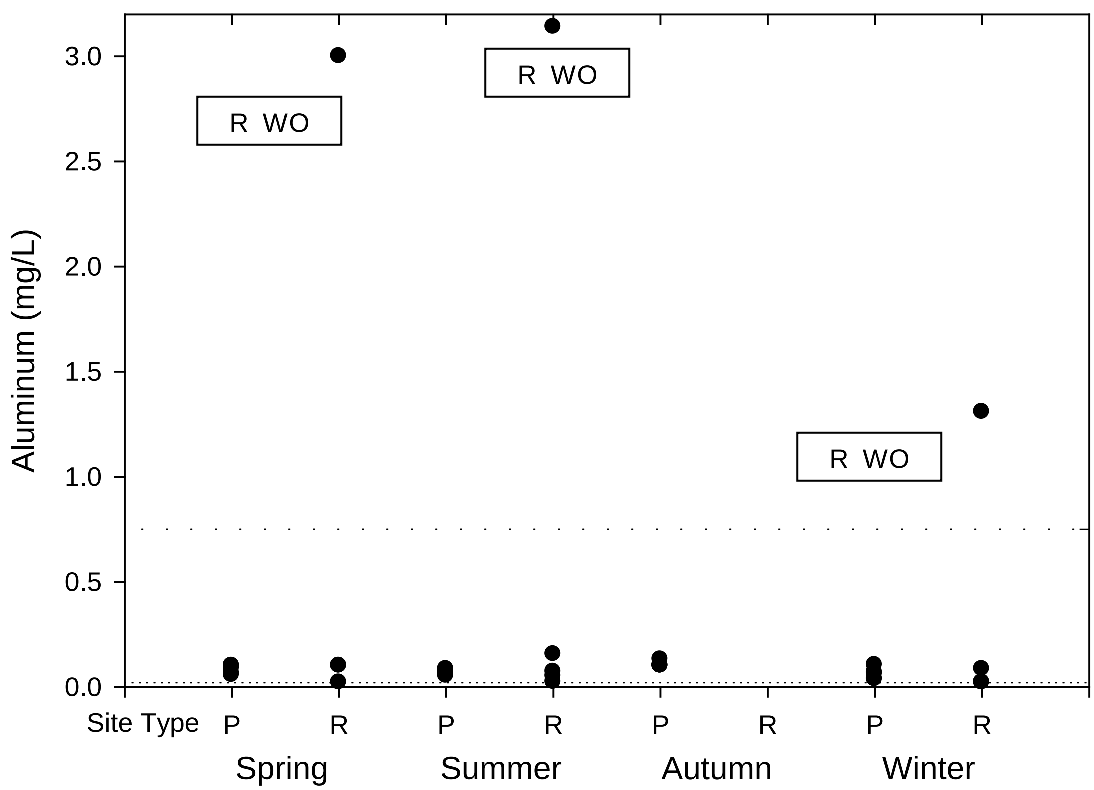

Figure 26. Seasonal aluminum (mg/L) for reclaimed mine perimeter channels and reference streams combined by site type. The WWF limit is 0.75 mg/L (dashed line). Method detection limits (MDL) were 0.021 mg/L (dotted line). Reference sites did not contain enough water for sampling in autumn.

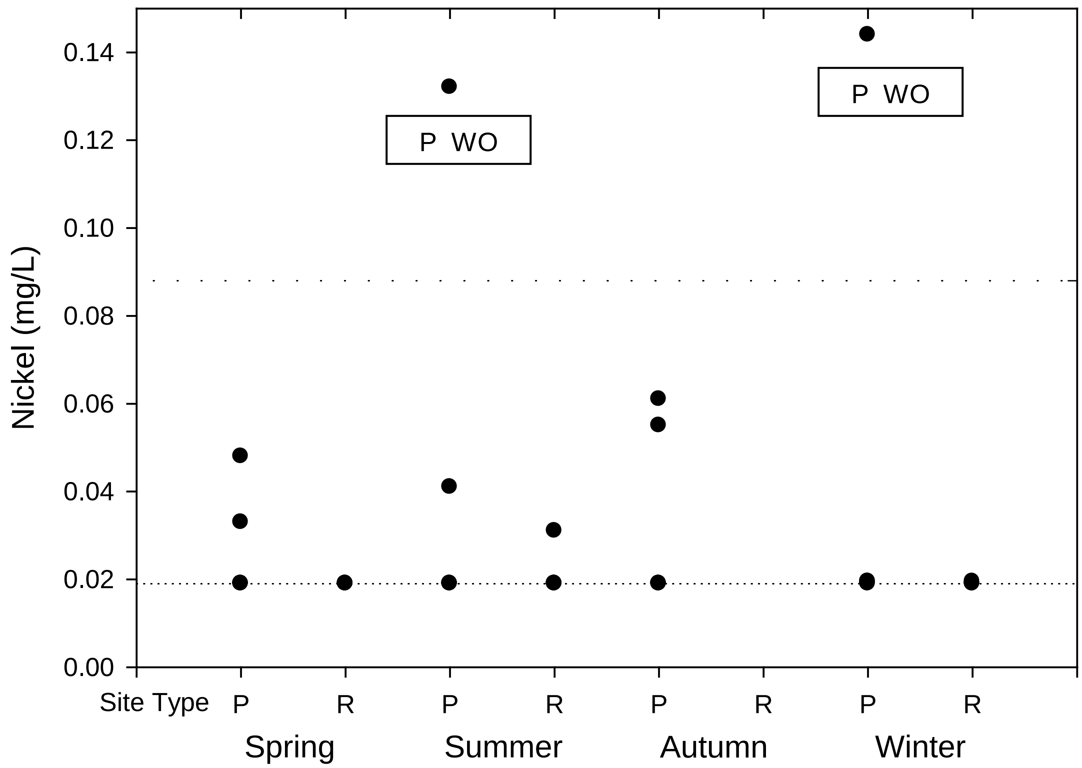

Figure 27. Seasonal nickel (mg/L) for reclaimed mine perimeter channels and reference streams combined by site type. Method detection limit (MDL) was 0.019 mg/L (dotted line). The WWF limit is 0.088 mg/L (dashed line). Reference sites did not contain enough water for sampling in autumn.

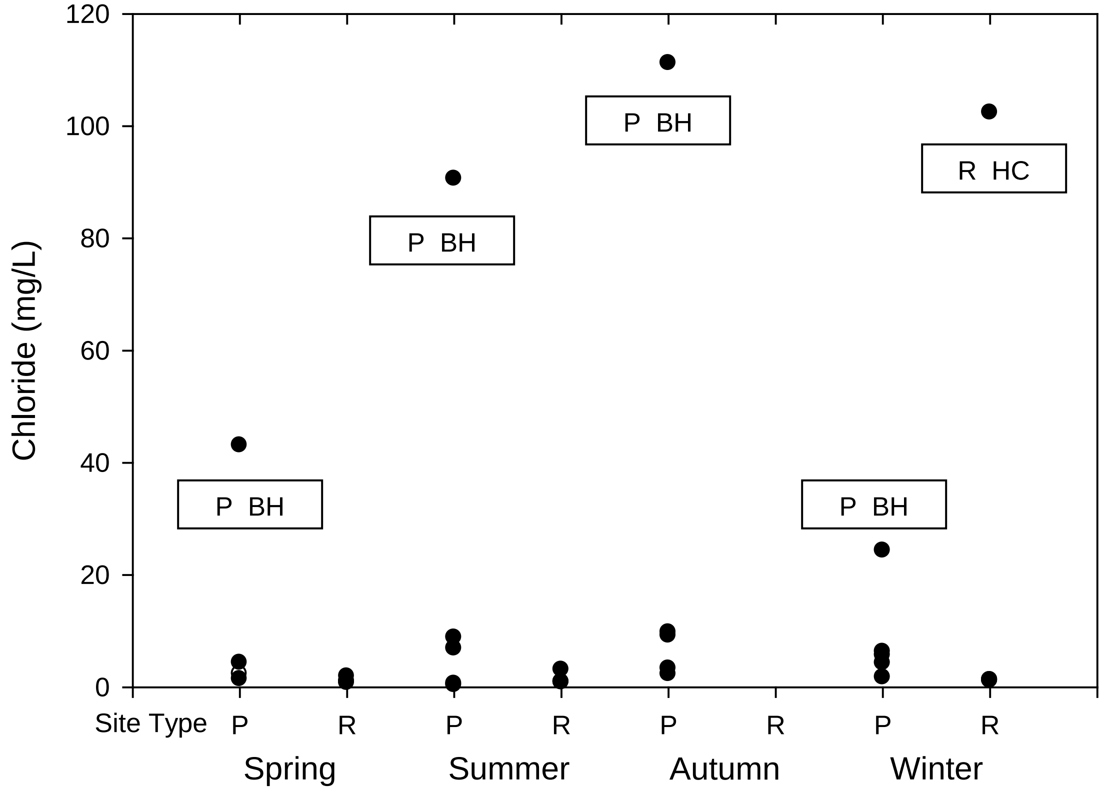

Figure 28. Seasonal chloride (mg/L) for reclaimed mine perimeter channels and reference streams combined by site type. Recommended limit for the protection of aquatic life is 600 mg/L. R_HC experienced a summer measure of 1070.73 mg/L (not shown) after disturbance. Reference sites did not contain enough water for sampling in autumn.

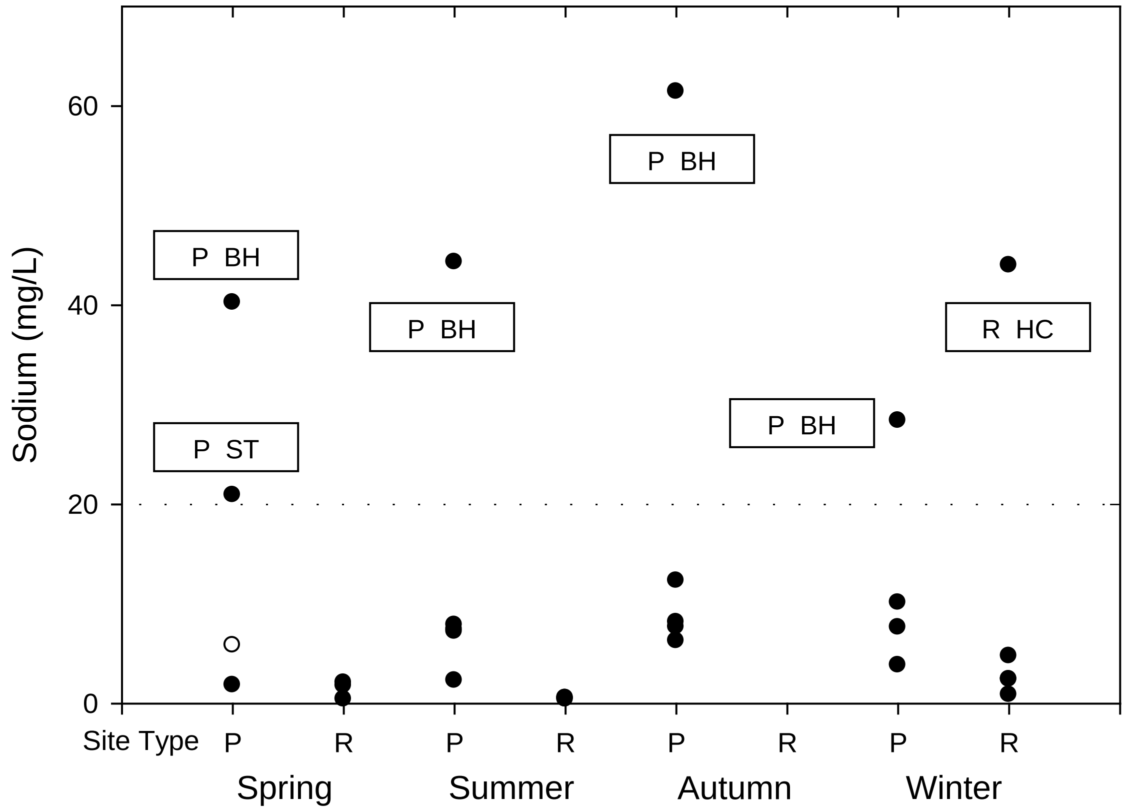

Figure 29. Seasonal sodium (mg/L) for reclaimed mine perimeter channels and reference streams combined by site type. Perimeter channels measured above 20 mg/L (dashed line). Summer R_HC measured 293.22 mg/L (not shown). Reference sites did not contain enough water for sampling in autumn.

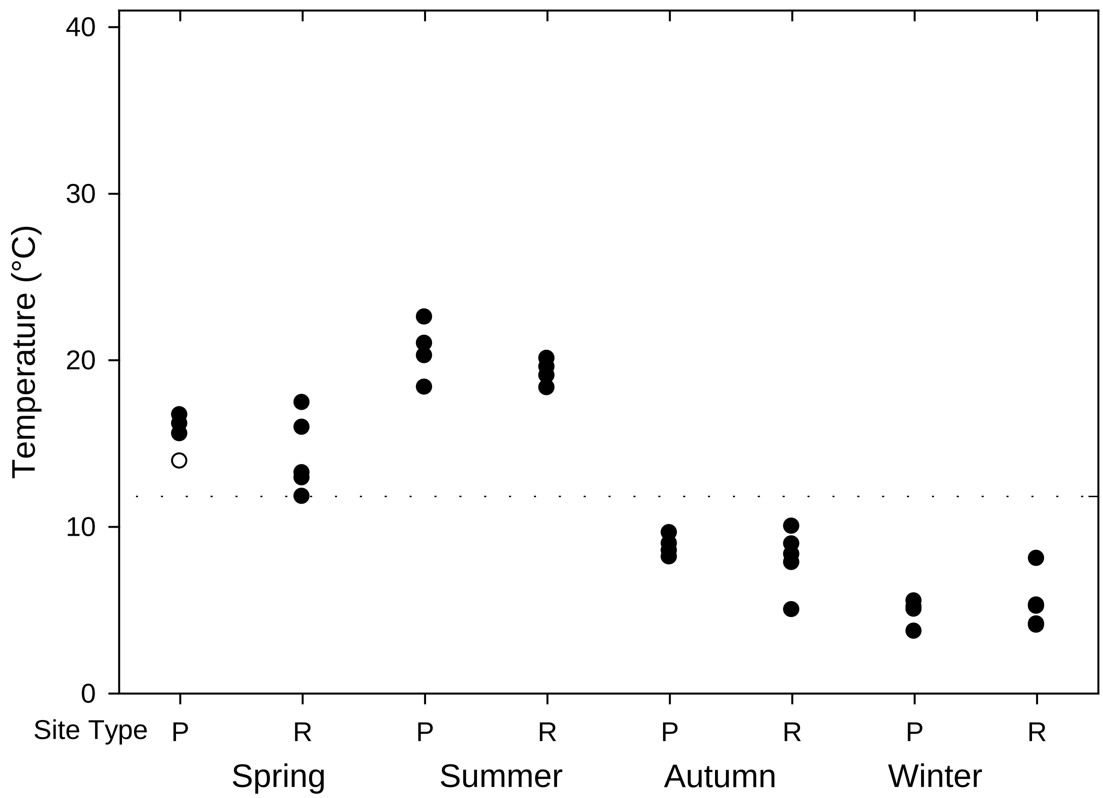

Figure 30. Mean daily temperature for reclaimed mine perimeter channels (P) and reference sites (R) during periods when streams contained water. The mean of these temperatures is 11.7 °C (dashed line).

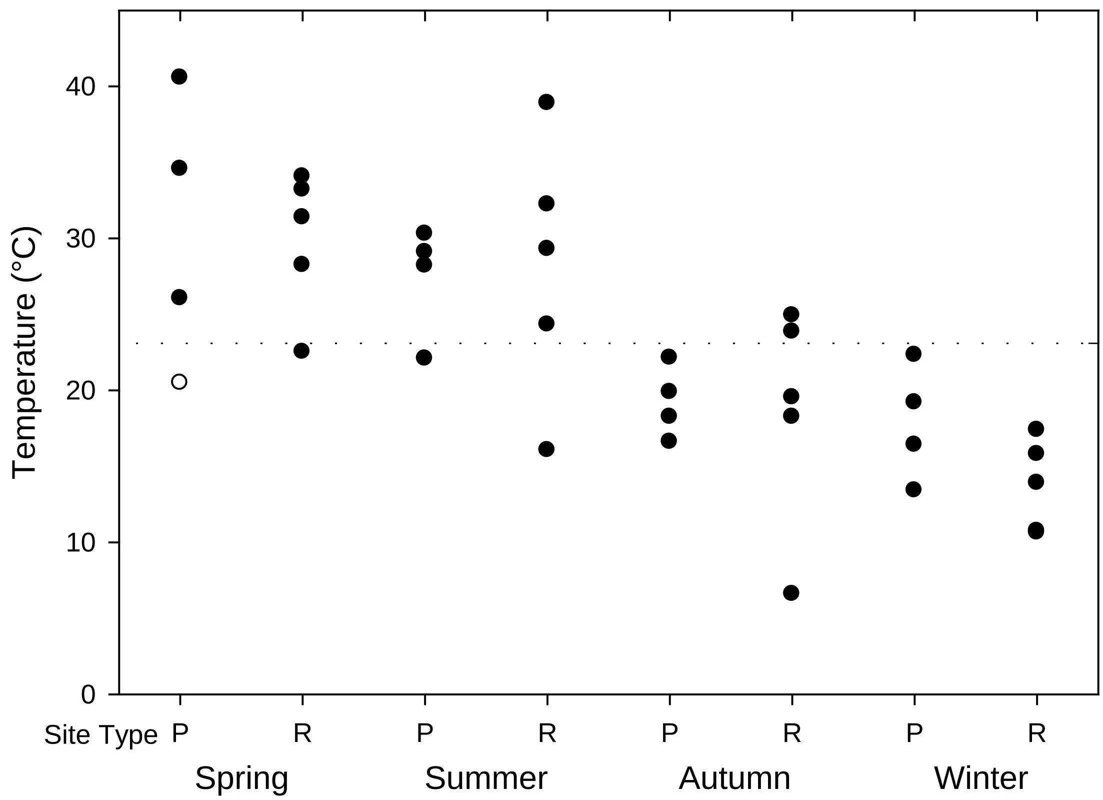

Figure 31. Maximum daily temperature for reclaimed mine perimeter channels (P) and reference sites (R) during periods when streams contained water. The mean of these temperatures is 22.9 °C (dashed line).

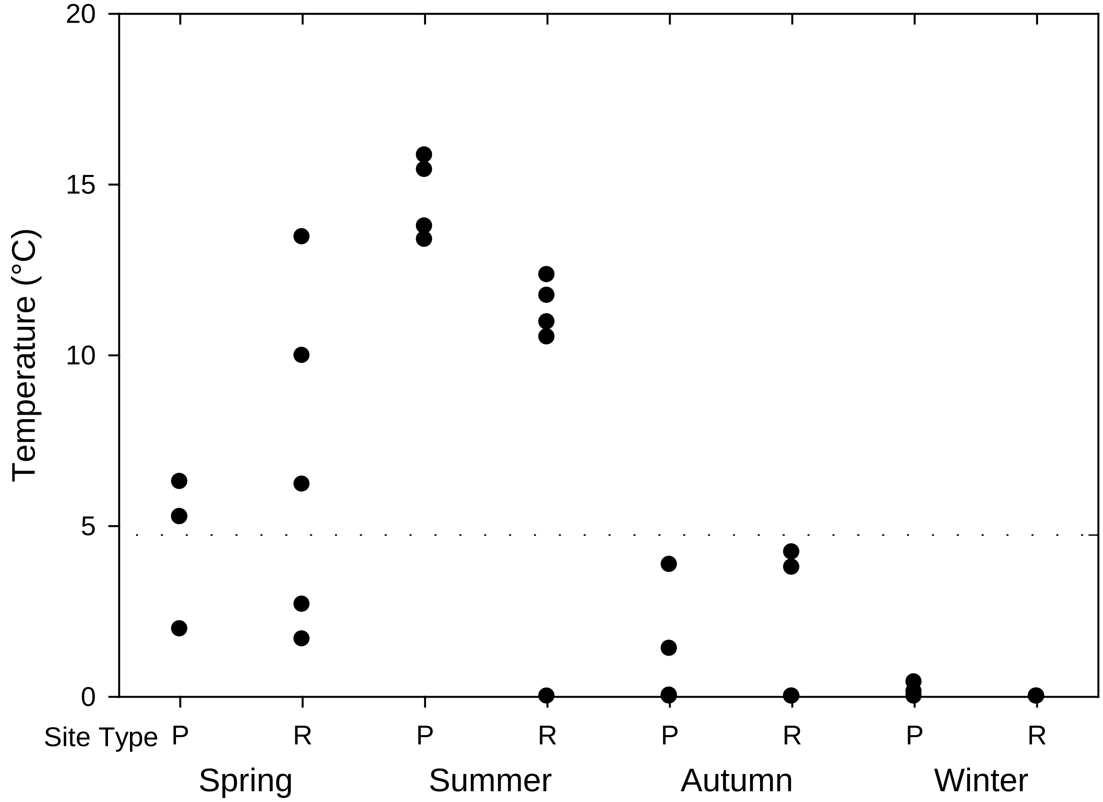

Figure 32. Minimum daily temperature for reclaimed mine perimeter channels (P) and reference sites (R) during periods when streams contained water. The mean of these temperatures is 4.7 °C (dashed line).

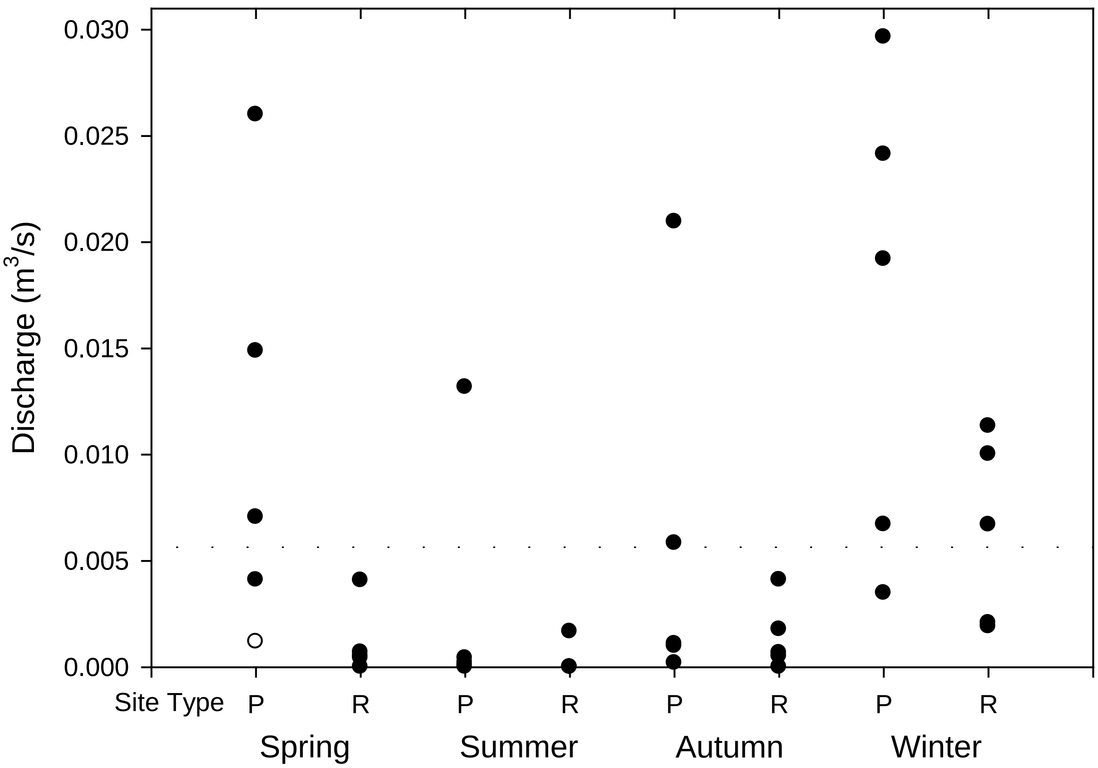

Figure 33. Seasonal discharge (m^3/s) for reclaimed mine perimeter channels and reference streams combined by site type. The mean discharge is 0.0057 m^3/s (dashed line).

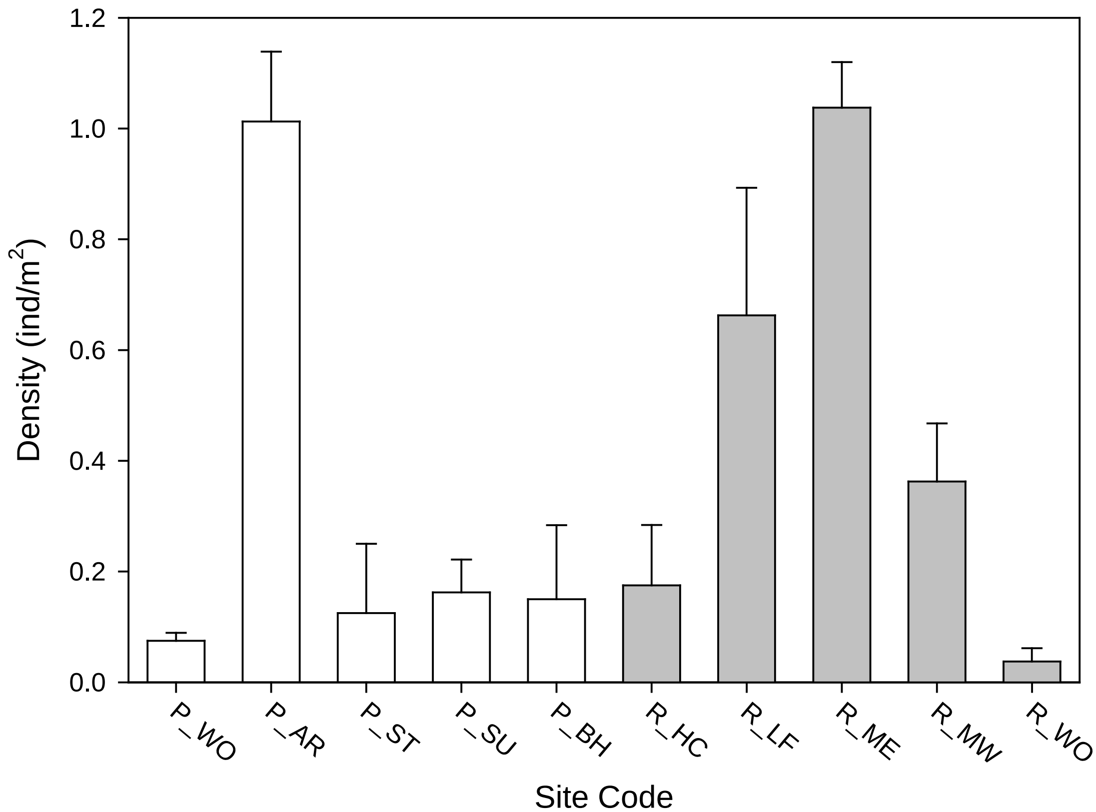

Figure 34. Mean amphibian density and standard error on reclaimed mine perimeter channels and reference sites determined from amphibian abundance surveys performed on four sample dates. Perimeter channel sites are listed in increasing age since reclamation. Perimeter channel sites are shown in black, and reference channel sites are shown in gray.

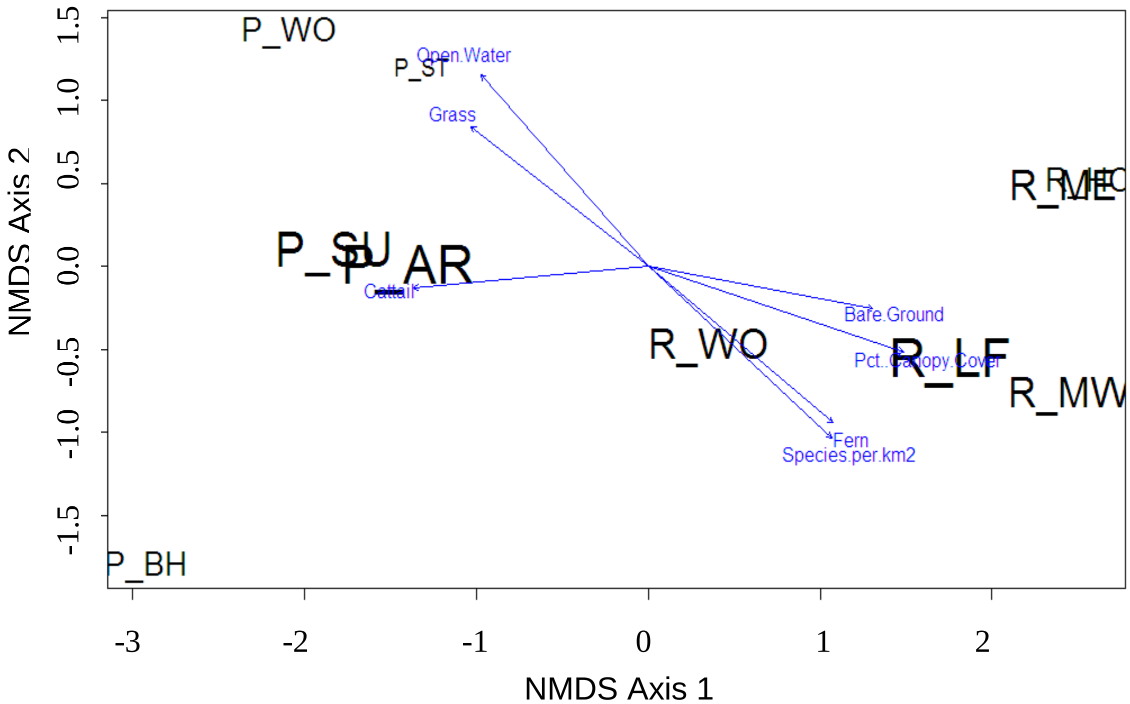

Figure 35. NMDS ordination analysis distinguishing site type by amphibian community data with overlaying significant vegetation vectors. Vegetation vectors include percent open water, grass, cattail, bare ground, percent canopy cover (Pct.Canopy.Cover), fern, and species per km^2. The direction of the vector indicates the direction of influence the vector has on determining community composition. The size of the character indicates the species richness of the site with larger characters indicating sites with greater richness.

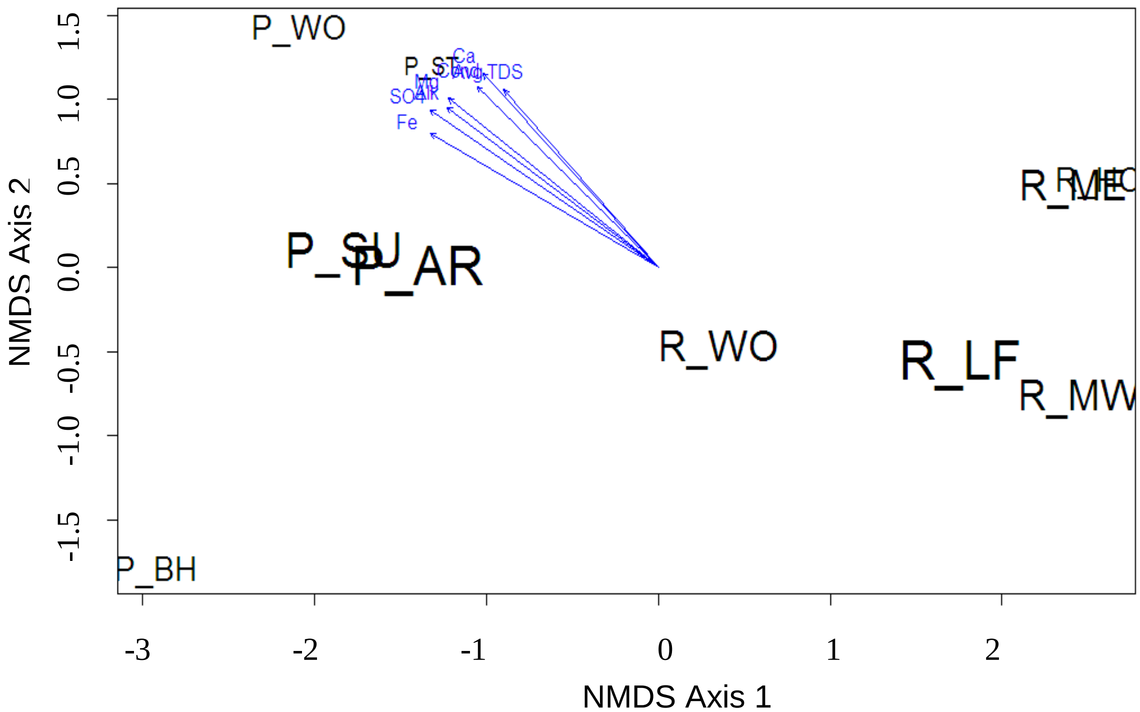

Figure 36. NMDS ordination analysis distinguishing site type by amphibian community data with overlaying significant water chemistry vectors. The direction of the vector indicates the direction of influence the vector has on determining community composition. Vector measures include mean conductivity (Cond), sulfate (SO_4), magnesium (Mg), mean total dissolved solids (Avg.TDS), calcium (Ca), alkalinity (Alk), and iron (Fe). The size of the character indicates the species richness of the site with larger characters indicating sites with greater richness.

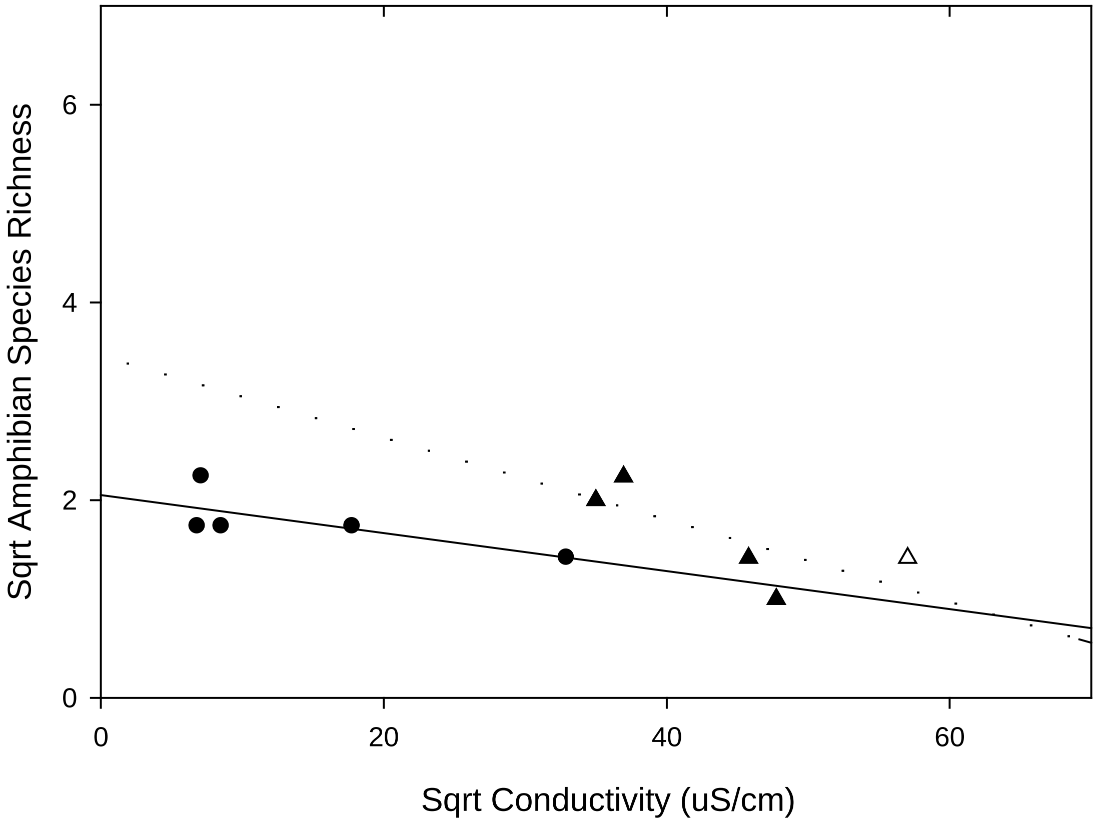

Figure 37. Mean conductivity versus amphibian species richness for four sampling periods. Reclaimed mine perimeter channel sites are represented by black triangles and dotted regression line. Reference sites are represented by black circles and solid regression line.

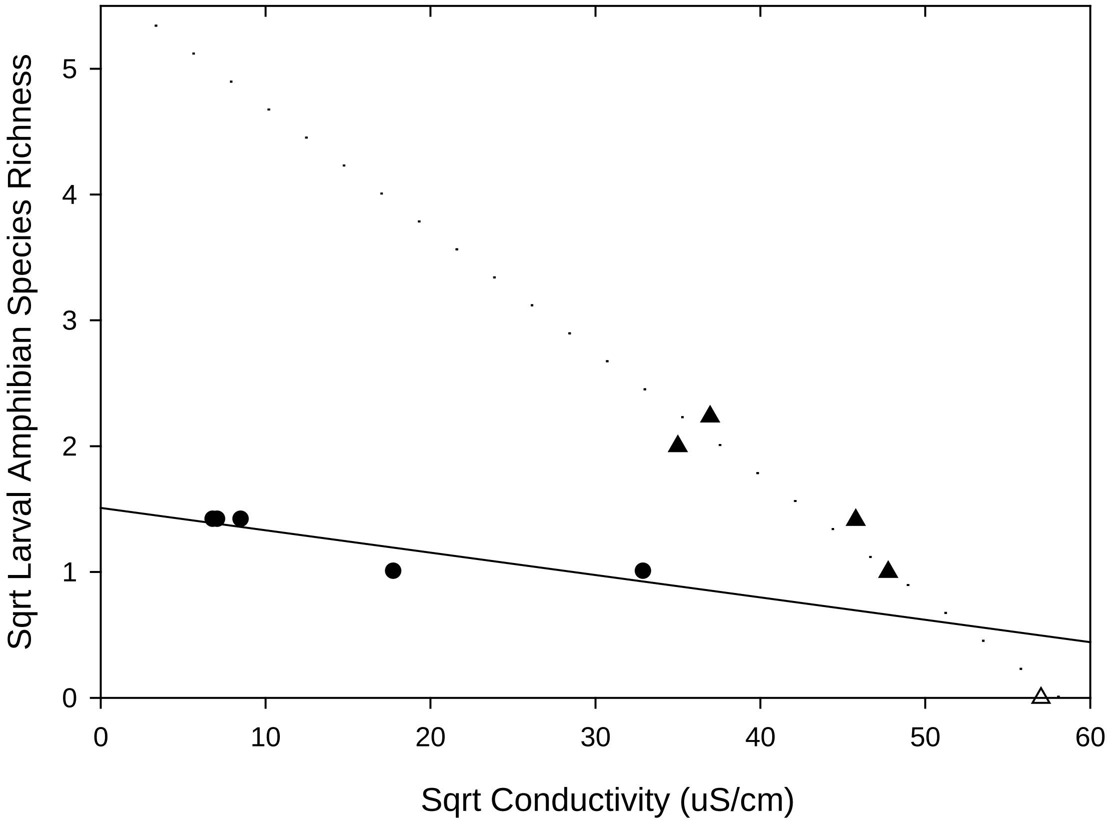

Figure 38. Mean conductivity versus the total number of larval amphibian species for four sampling periods. Reclaimed mine perimeter channel sites are represented by black triangles and dotted regression line. Reference sites are represented by black circles and solid regression line.

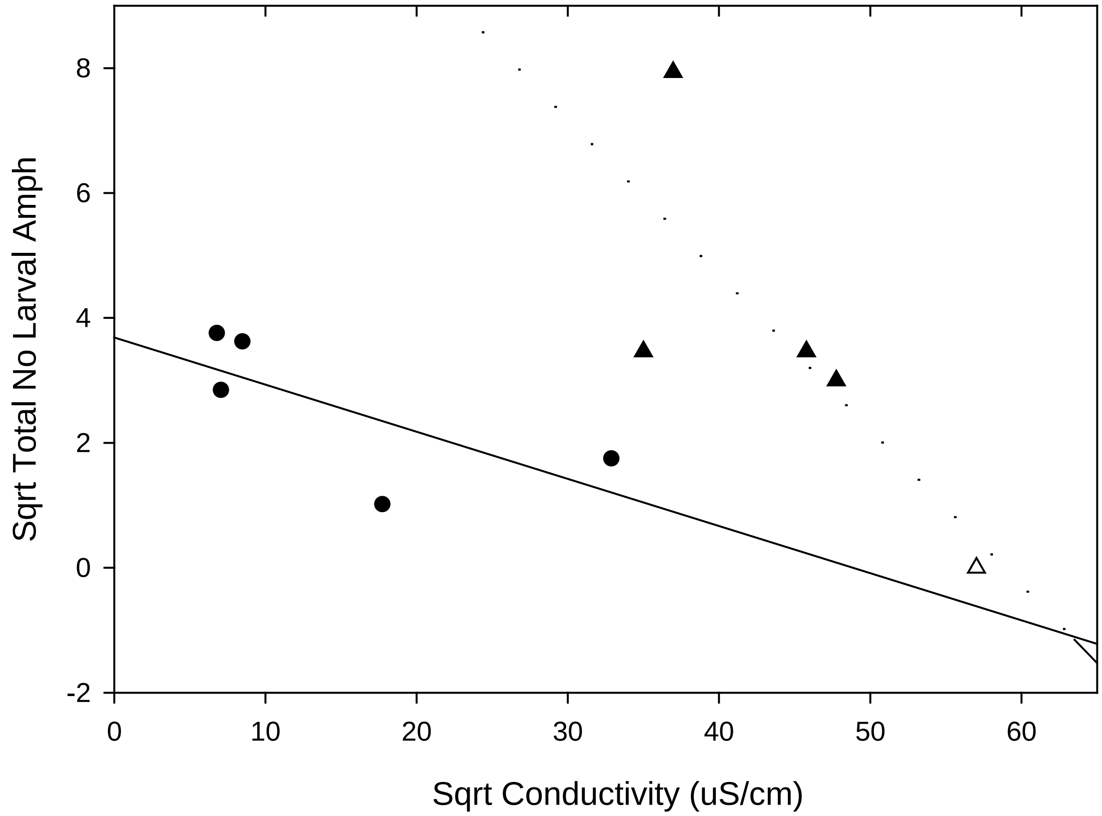

Figure 39. Mean conductivity versus the total number of larval amphibians captured during four amphibian sampling periods. Reclaimed mine perimeter channel sites are represented by black triangles and dotted regression line. Reference sites are represented by black circles and solid regression line.

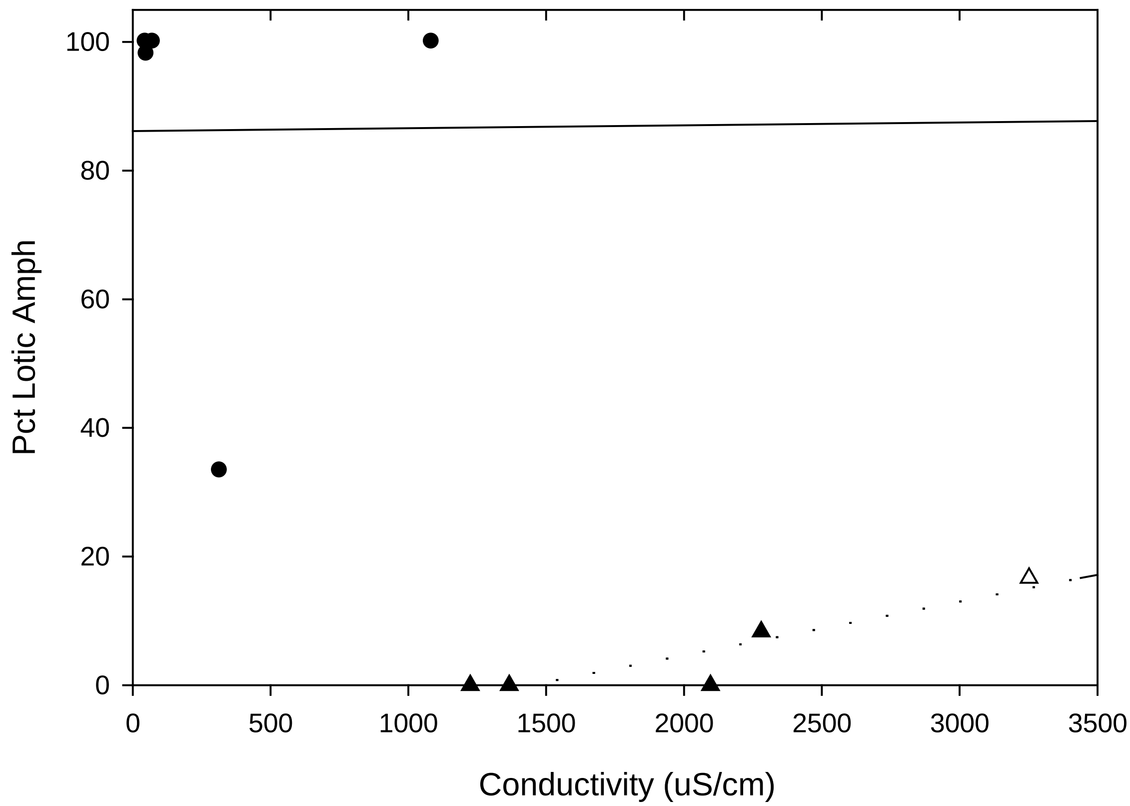

Figure 40. Mean conductivity versus mean percentage of amphibian species present that utilize lotic habitat. Reclaimed mine perimeter channel sites are represented by black triangles and dotted regression line. Reference sites are represented by black circles and solid regression line.

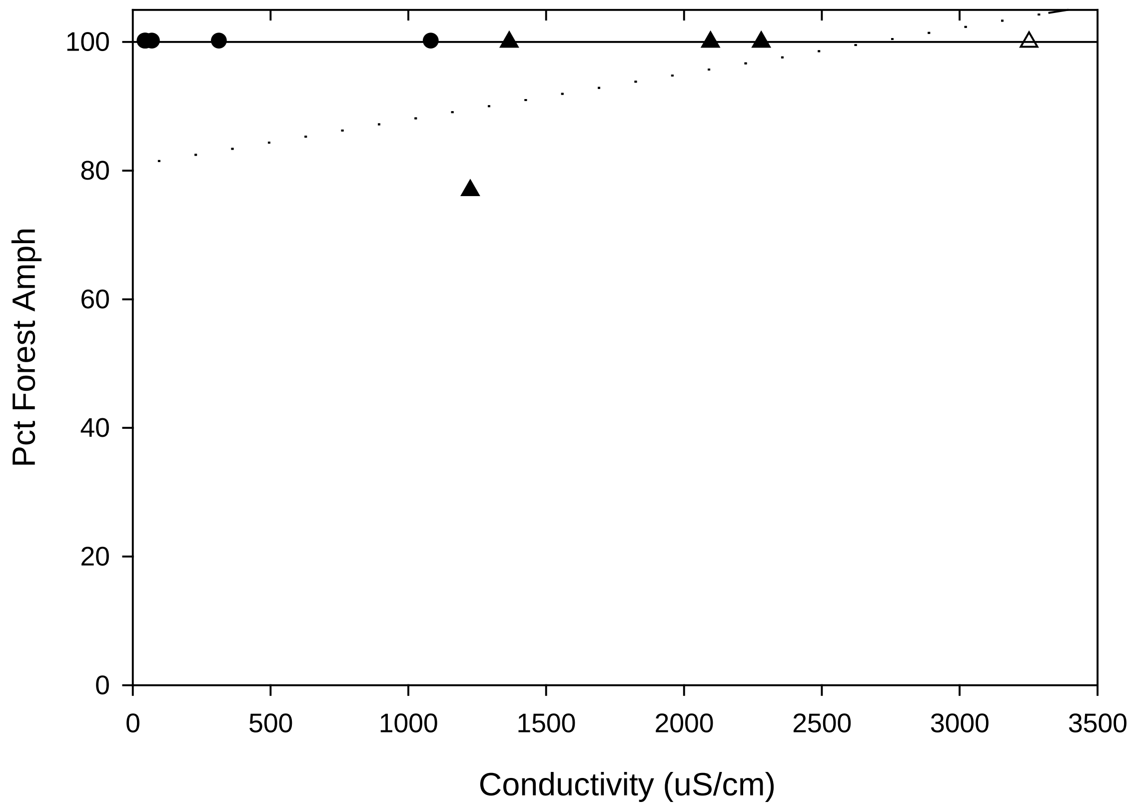

Figure 41. Mean conductivity versus the percentage of amphibian species present, during four sampling periods, that utilize forest habitat. Reclaimed mine perimeter channel sites are represented by black triangles and dotted regression line. Reference sites are represented by black circles and solid regression line.

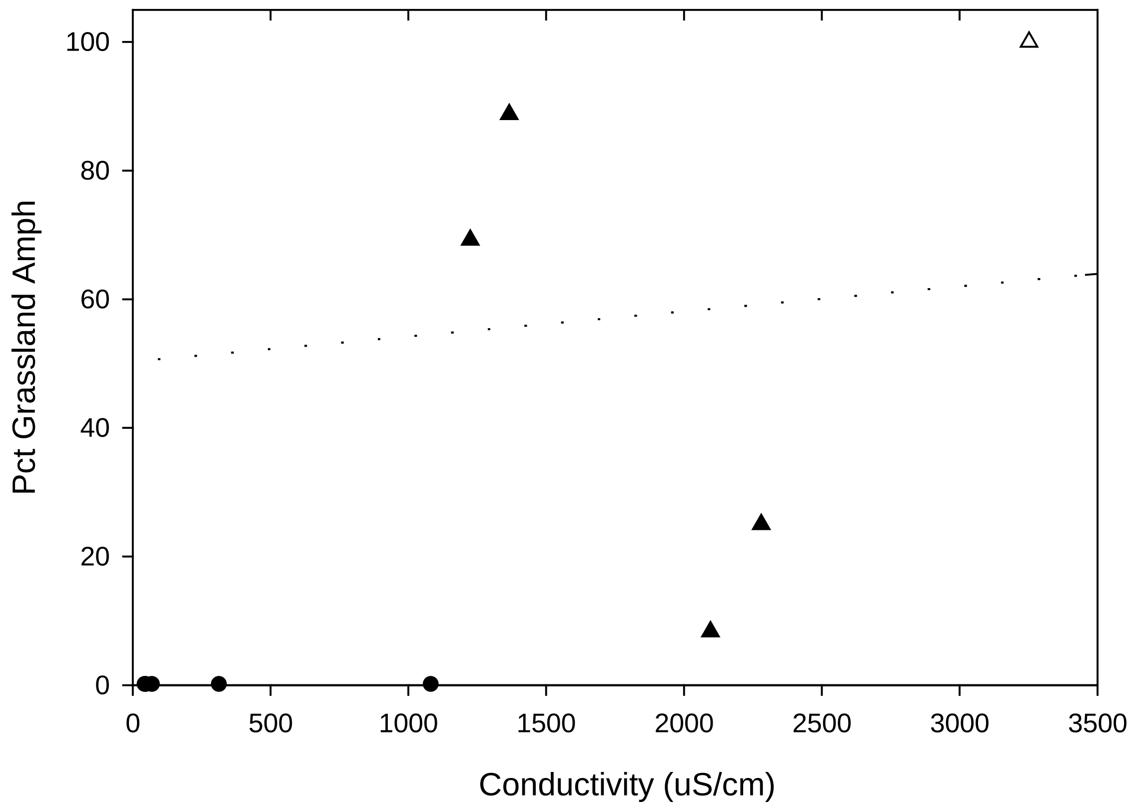

Figure 42. Mean conductivity versus the percentage of amphibian species present that utilize grassland habitat. Reclaimed mine perimeter channel sites are represented by black triangles and dotted regression line. Reference sites are represented by black circles and solid regression line.

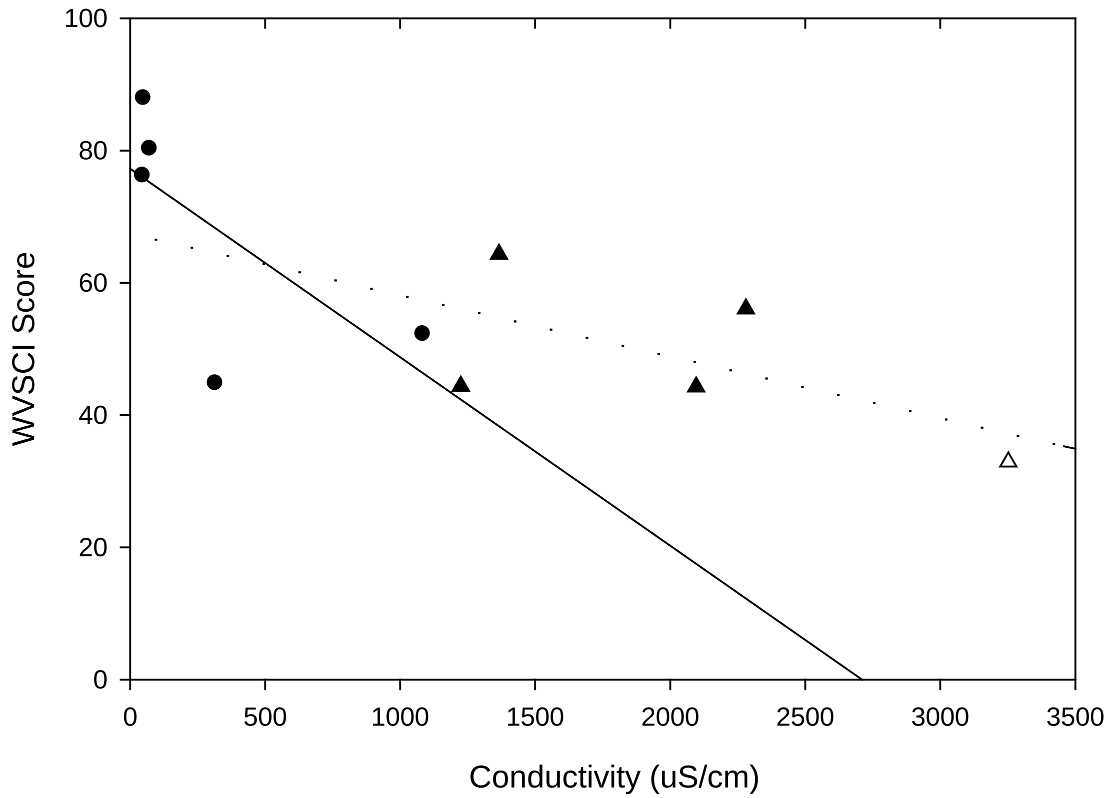

Figure 43. Mean conductivity versus WVSCI score for the spring 2008 sampling period. Reclaimed mine perimeter channel sites are represented by black triangles and dotted regression line. Reference sites are represented by black circles and solid regression line.

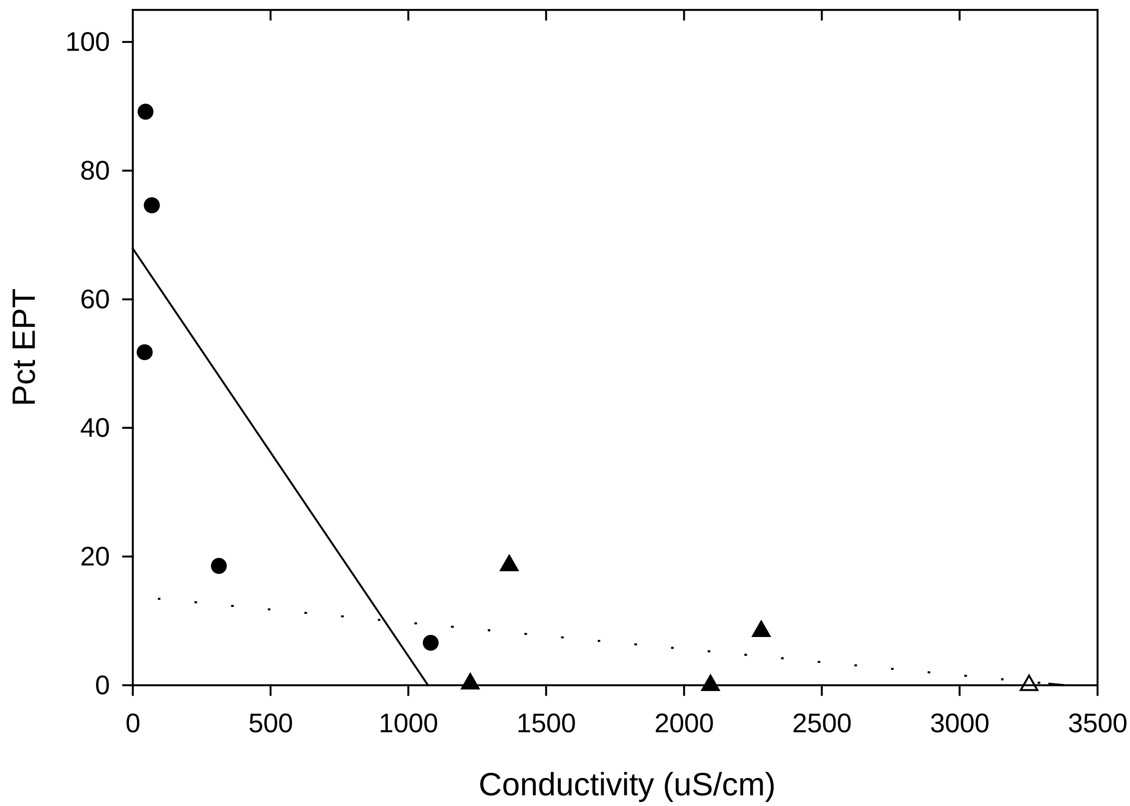

Figure 44. Mean conductivity versus percent EPT for the spring 2008 sampling period. Reclaimed mine perimeter channel sites are represented by black triangles and dotted regression line. Reference sites are represented by black circles and solid regression line.

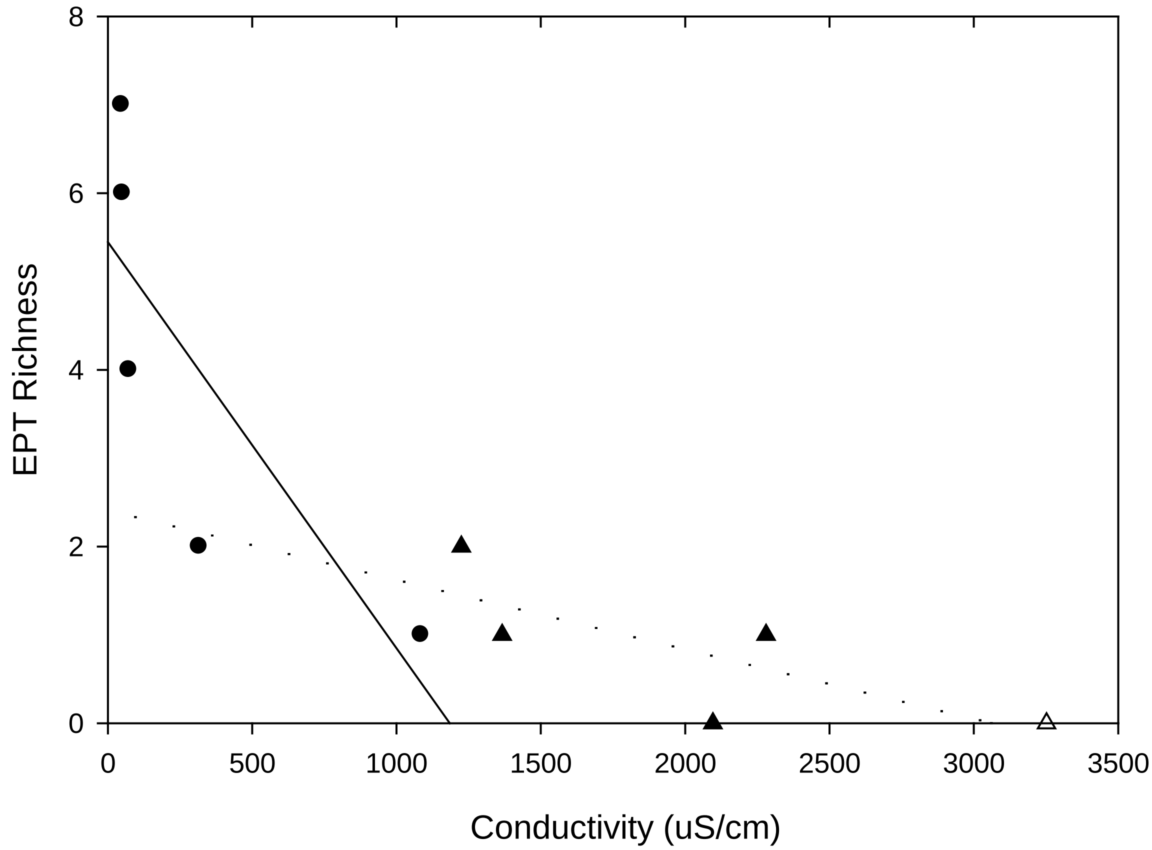

Figure 45. Mean conductivity versus EPT species richness for the spring 2008 sampling period. Reclaimed mine perimeter channel sites are represented by black triangles and dotted regression line. Reference sites are represented by black circles and solid regression line.

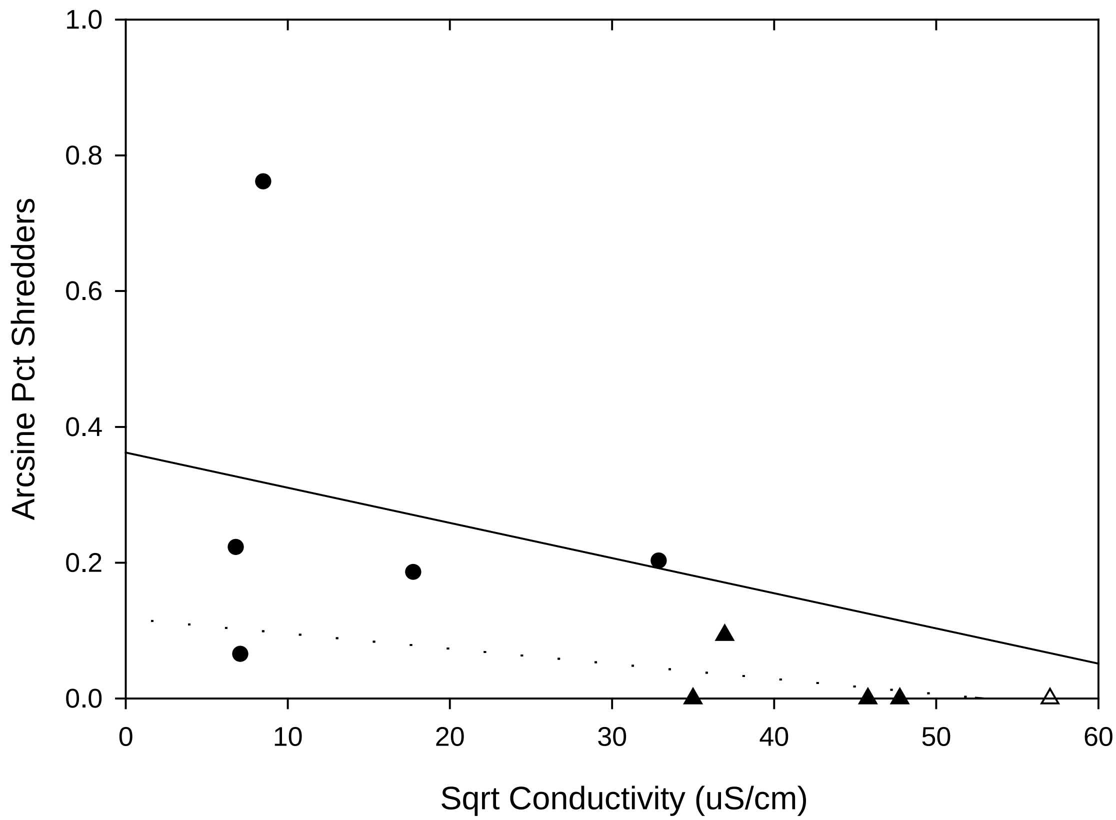

Figure 46. Mean conductivity versus percentage of shredders for the spring 2008 sampling period. Reclaimed mine perimeter channel sites are represented by black triangles and dotted regression line. Reference sites are represented by black circles and solid regression line

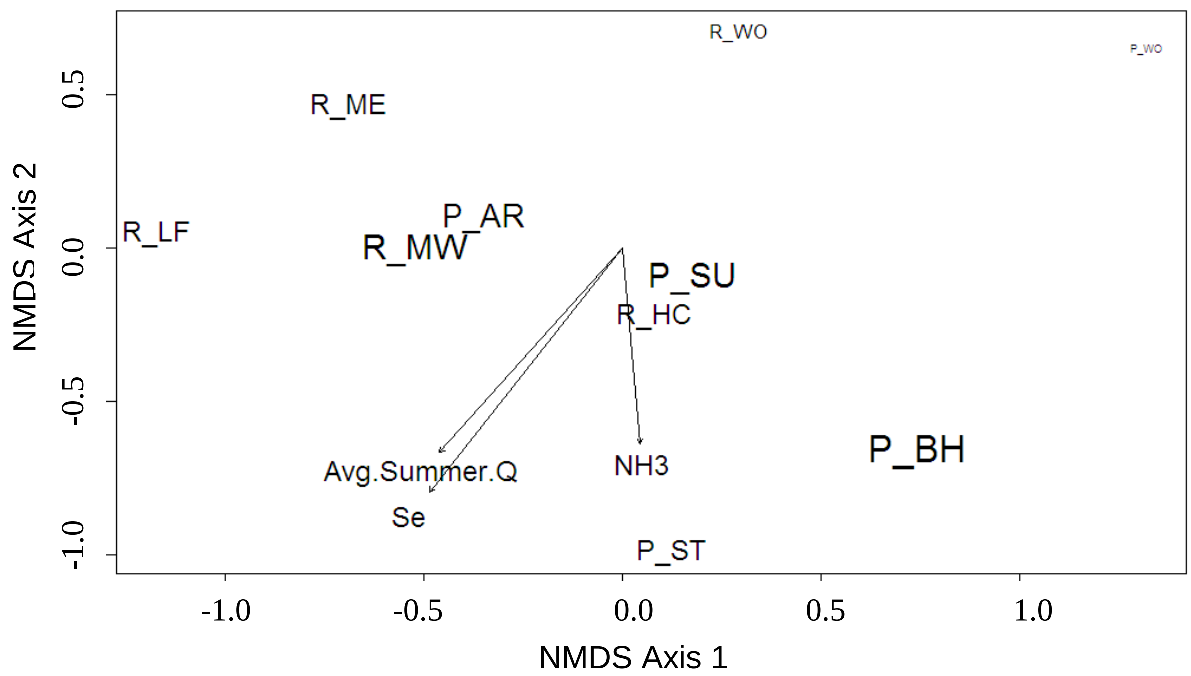

Figure 47. NMDS ordination analysis distinguishing site type by macroinvertebrate community data with overlaying significant environmental vectors. The direction of the vector indicates the direction of influence the vector has on determining community composition. Vector measures include NH_3, mean summer discharge (Avg.Summer.Q), and selenium (Se). The size of the site character indicates the family richness of the site with larger characters indicating sites with greater richness.

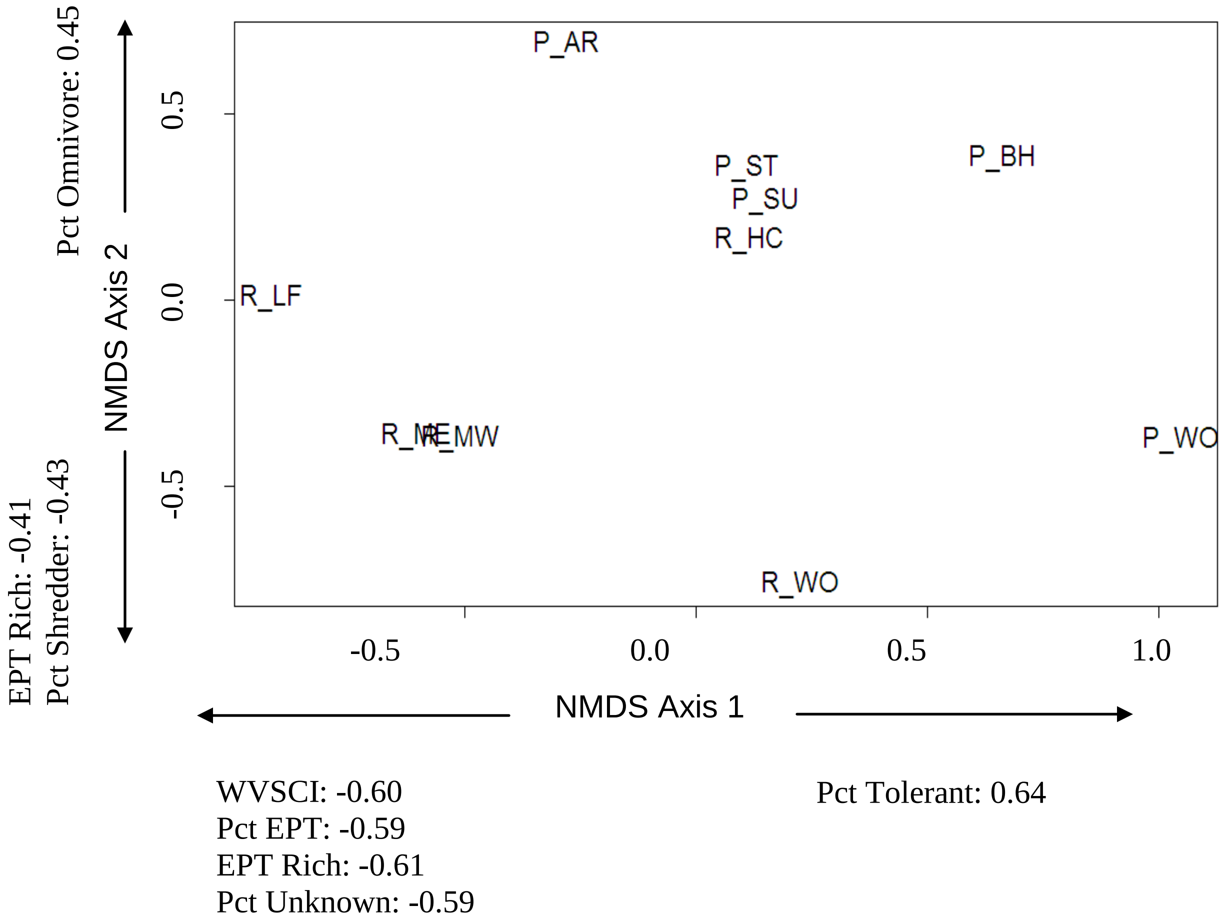

Figure 48. NMDS ordination analysis distinguishing site type by macroinvertebrate community data with Spearman rank correlations are annotated along each axis. The size of the site character indicates the species richness of the site with larger characters indicating sites with greater richness.

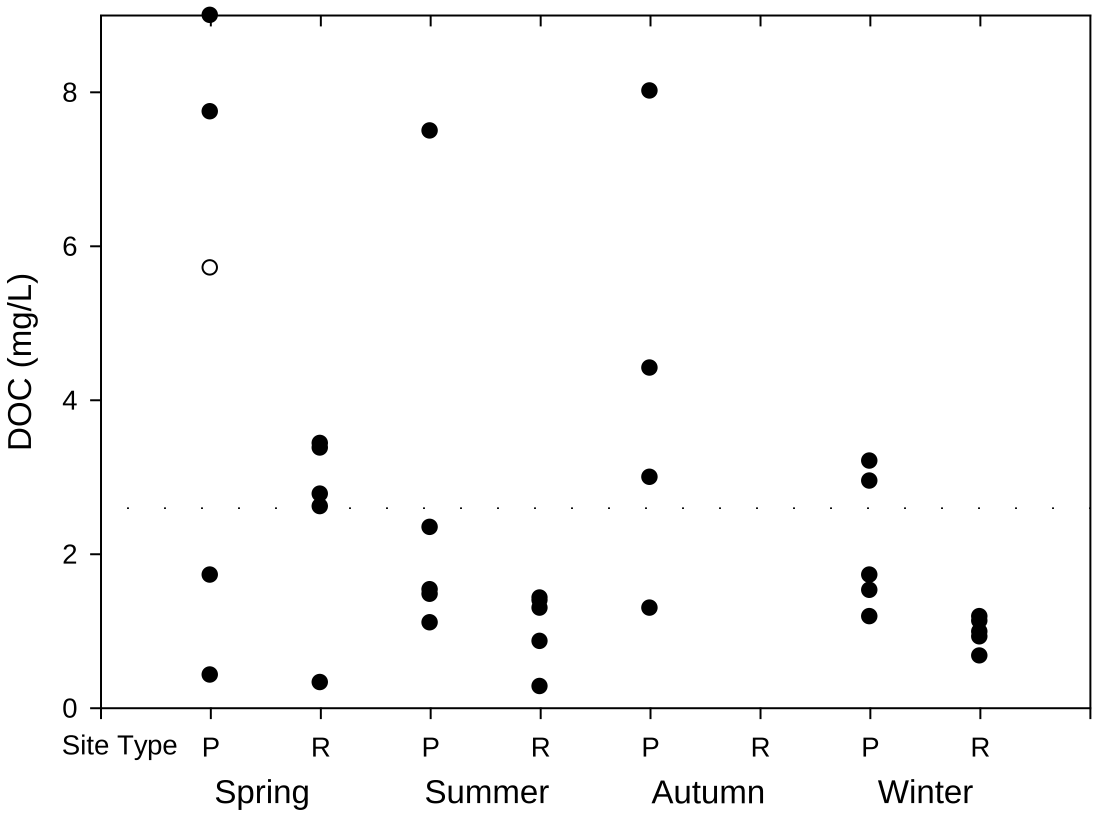

Figure 49. Seasonal DOC measurements for reclaimed mine perimeter channels and reference sites combined by site type. Mean DOC is 2.60 mg/L (dashed line). Reference sites did not contain enough water for autumn sampling.

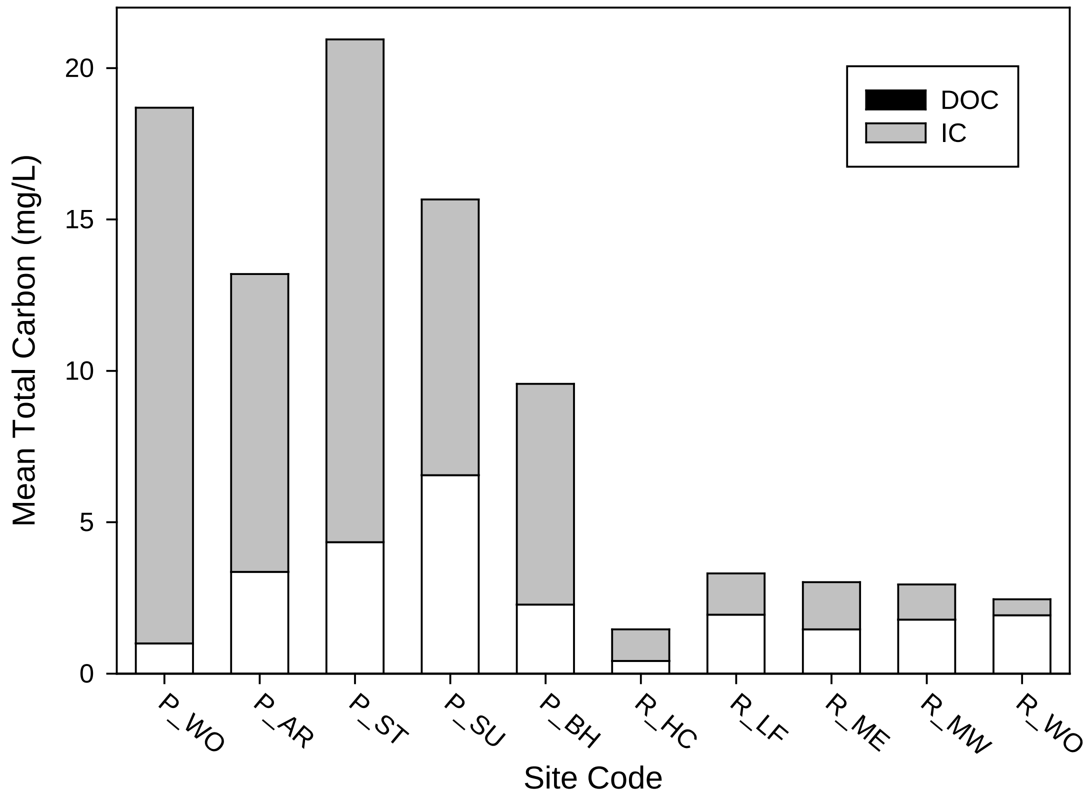

Figure 50. Mean total carbon for reclaimed mine perimeter channels and reference sites given by concentration of dissolved organic carbon (DOC) and inorganic carbon (IC). Perimeter channel sites are listed in increasing age since reclamation.

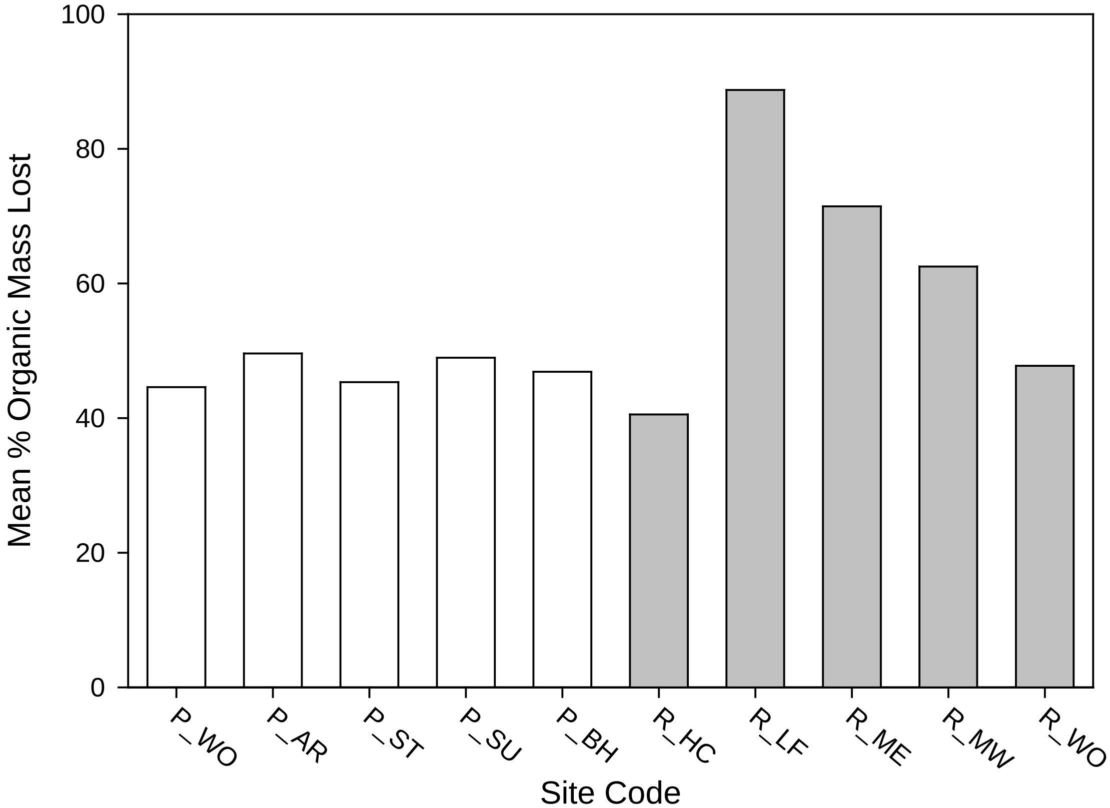

Figure 51. Mean percent organic mass lost from original leaf litter inputs after ~325 days of exposure on reclaimed mine perimeter channels and reference sites. Perimeter channel sites are listed in order of increasing age since reclamation. Perimeter channel sites are shown in black and reference channel sites are shown in gray.

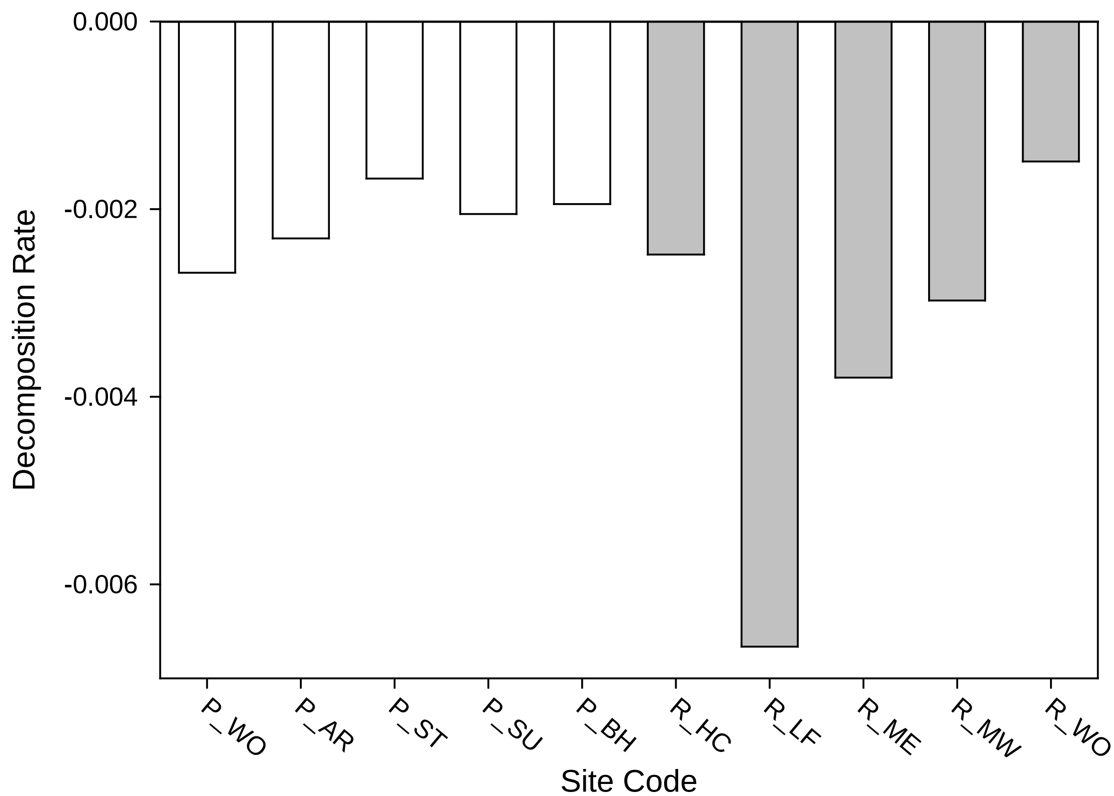

Figure 52. Decomposition rate (-k) of *Quercus palustris* (pin oak) leaf litter on reclaimed mine perimeter channels and reference streams after ~325 days of exposure. Perimeter channel sites are listed in increasing age since reclamation. Perimeter channel sites are shown in black and reference channel sites are shown in gray.

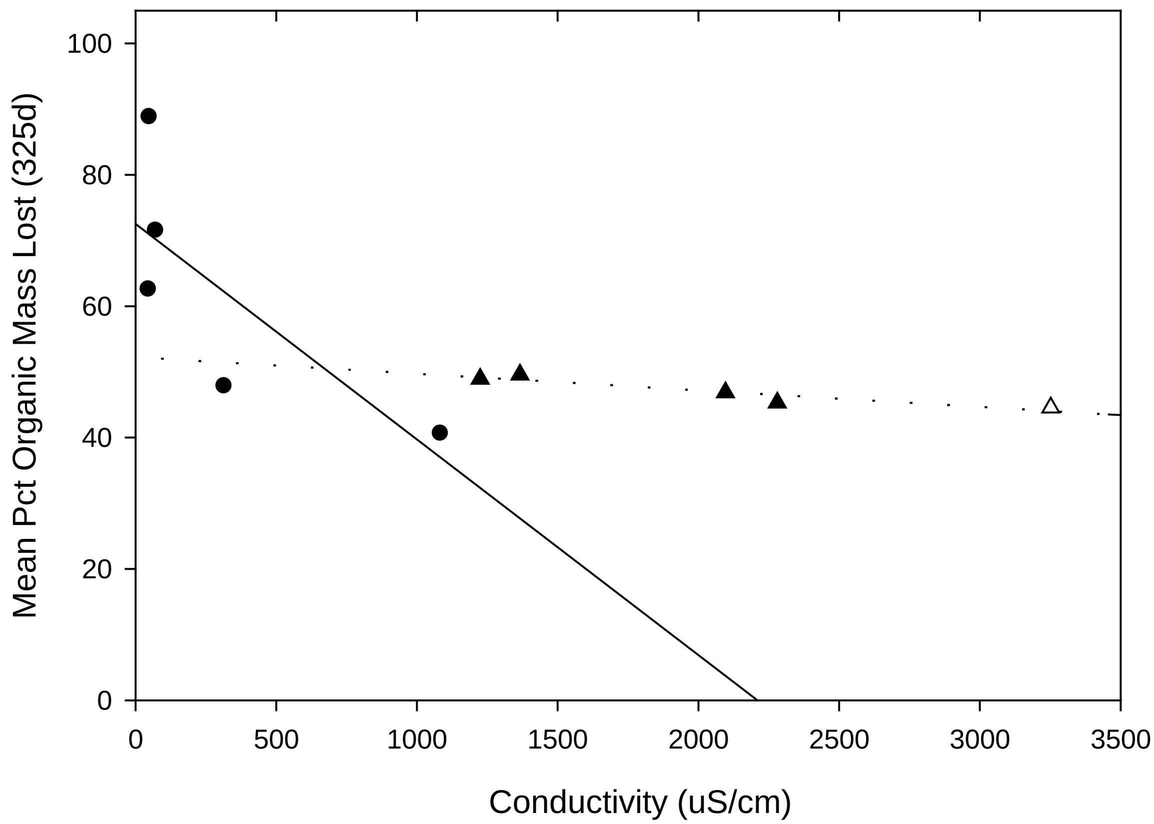

Figure 53. Mean conductivity versus mean percent organic mass lost from leaf litter packs after 325 days. Reclaimed mine perimeter channel sites are represented by black triangles and dotted regression line. Reference sites are represented by black circles and solid regression line.

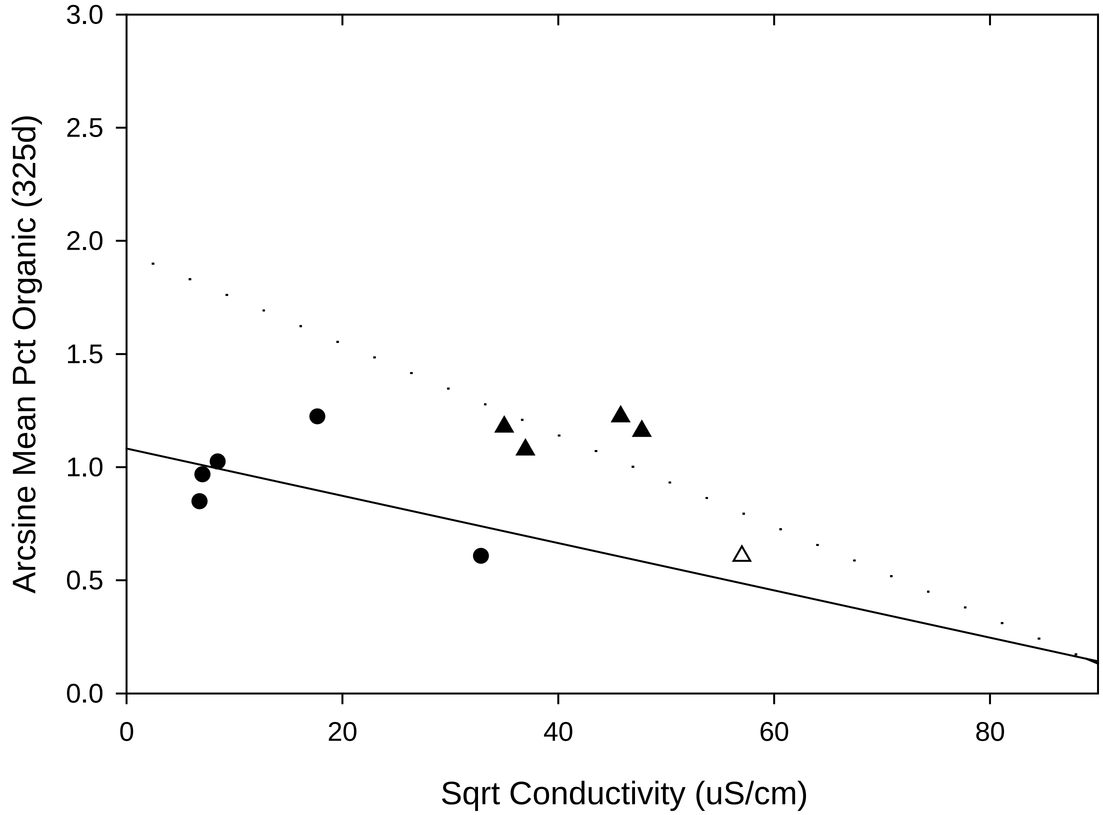

Figure 54. Mean conductivity versus mean percent organic matter composition in leaf litter packs after 325 days. Reclaimed mine perimeter channel sites are represented by black triangles and dotted regression line. Reference sites are represented by black circles and solid regression line.

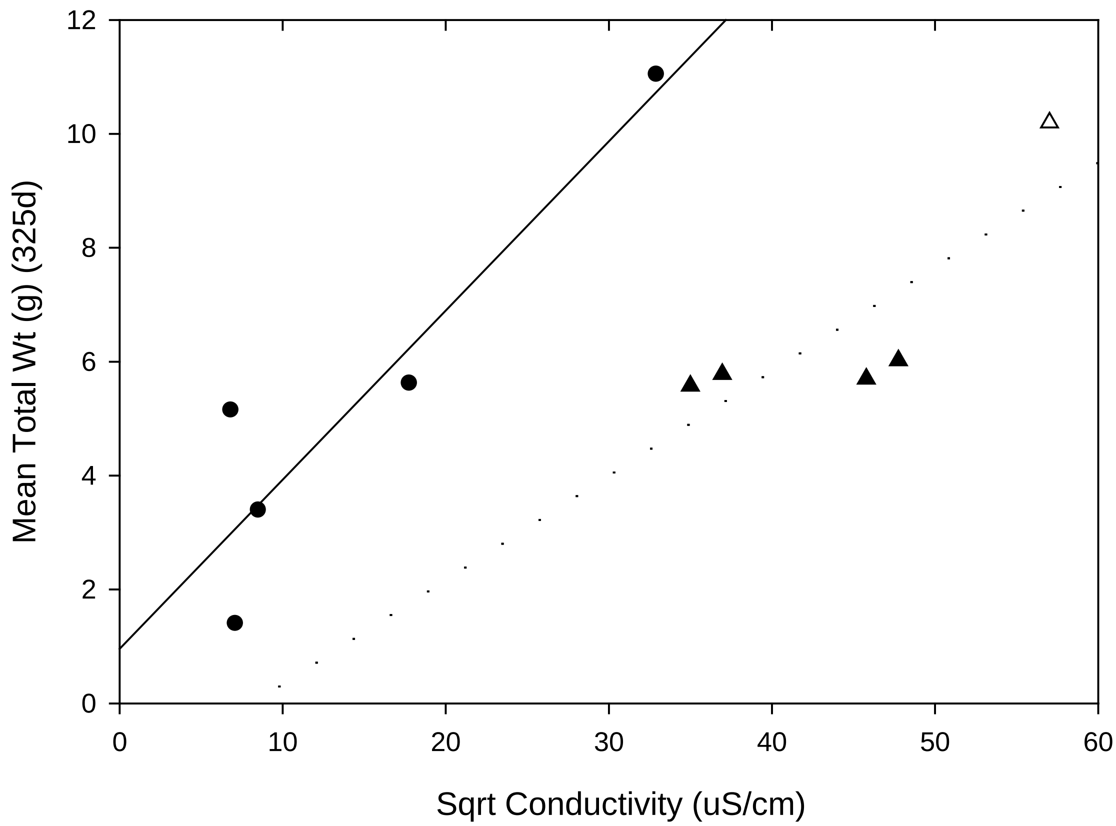

Figure 55. Mean conductivity versus mean total weight of leaf litter packs after ~325 days. Reclaimed mine perimeter channel sites are represented by black triangles and dotted regression line. Reference sites are represented by black circles and solid regression line.

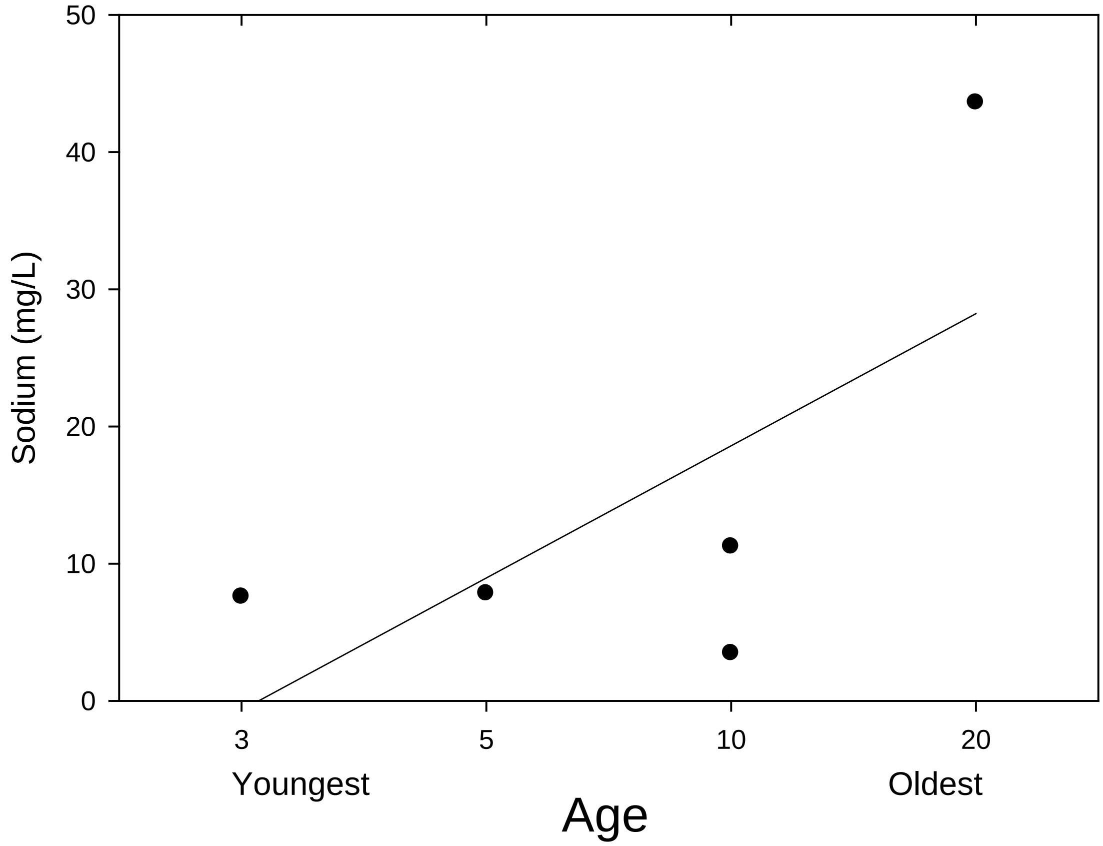

Figure 56. Mean sodium (mg/L) levels for reclaimed mine perimeter channel sites in order of age since reclamation. Confidence interval for reference sites (-16.7 – 71.1 mg/L) is not shown.

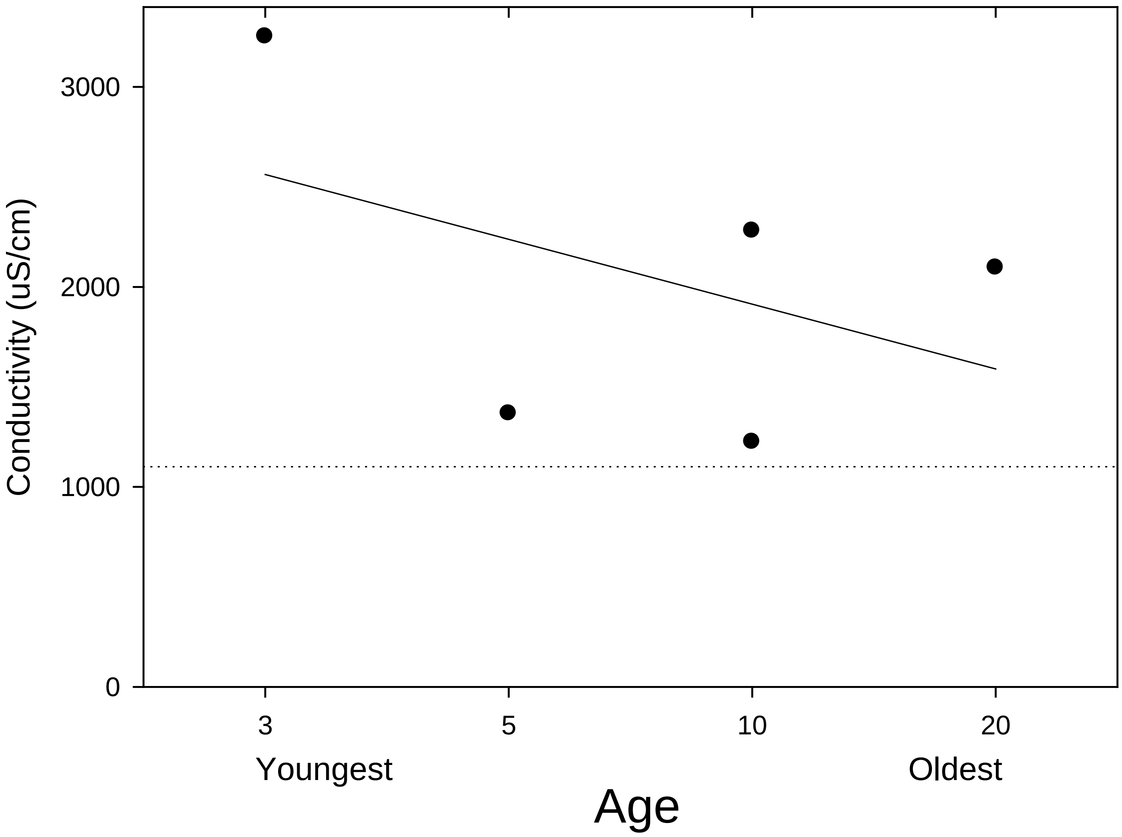

Figure 57. Mean conductivity (µS/cm) levels for reclaimed mine perimeter channel sites in order of age since reclamation. Confidence interval for reference sites (-179 – 1101 µS/cm) is shown (dotted line).

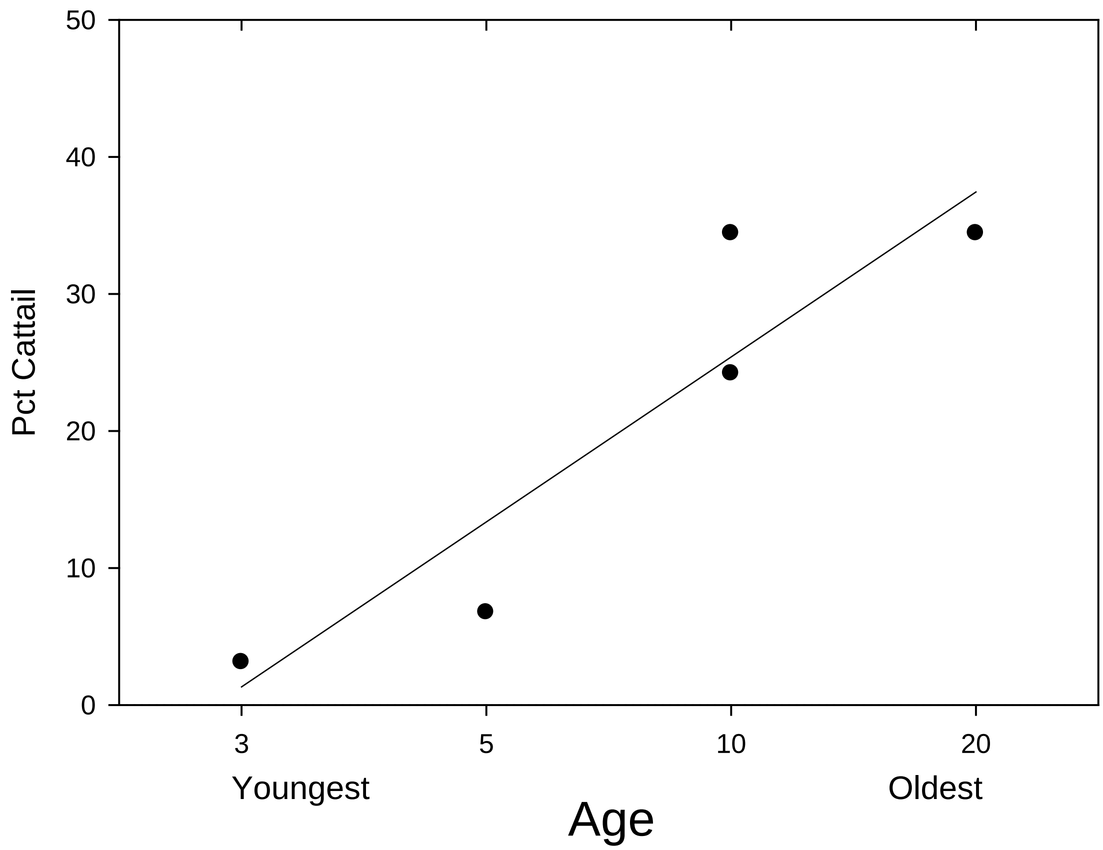

Figure 58. Percent cattail for reclaimed mine perimeter channel sites in order of age since reclamation. Confidence interval for reference sites was zero.

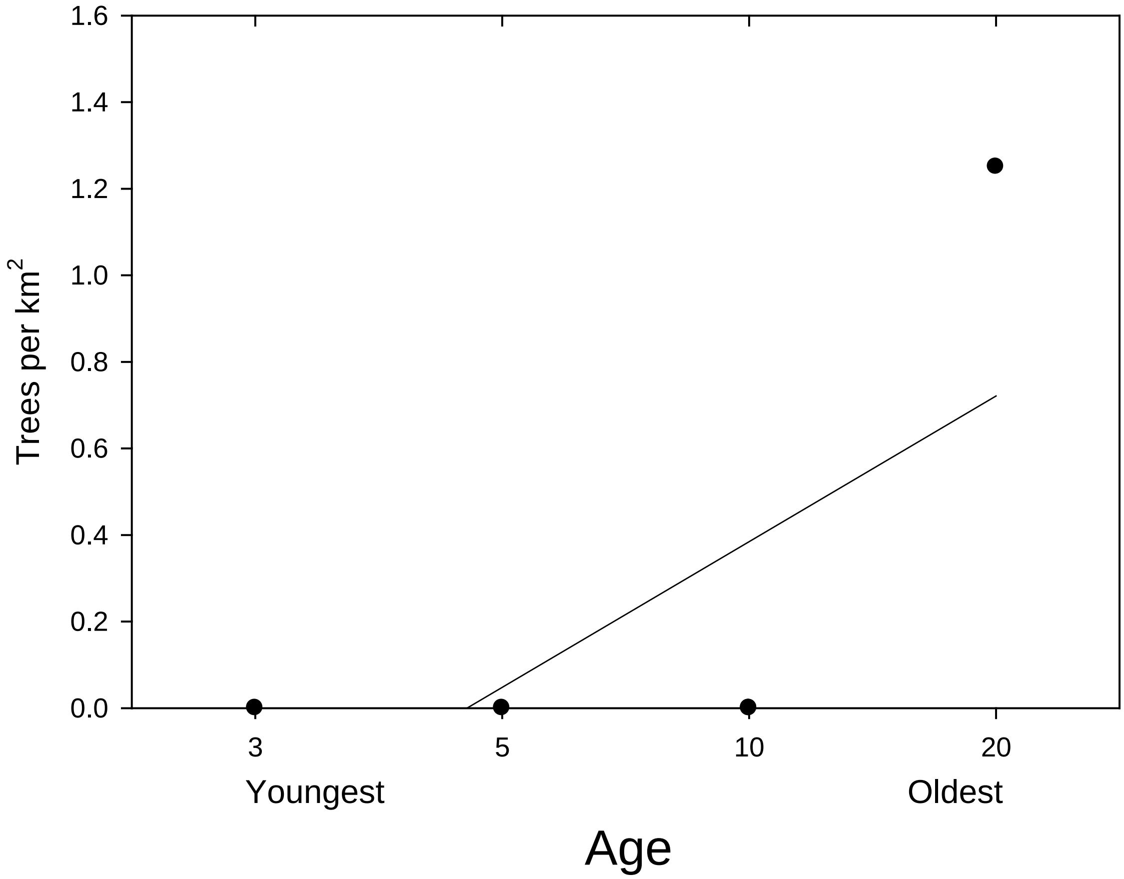

Figure 59. Trees per km^2 for reclaimed mine perimeter channel sites in order of age since reclamation. Confidence interval for reference sites (4.8 – 13.5) is not shown.

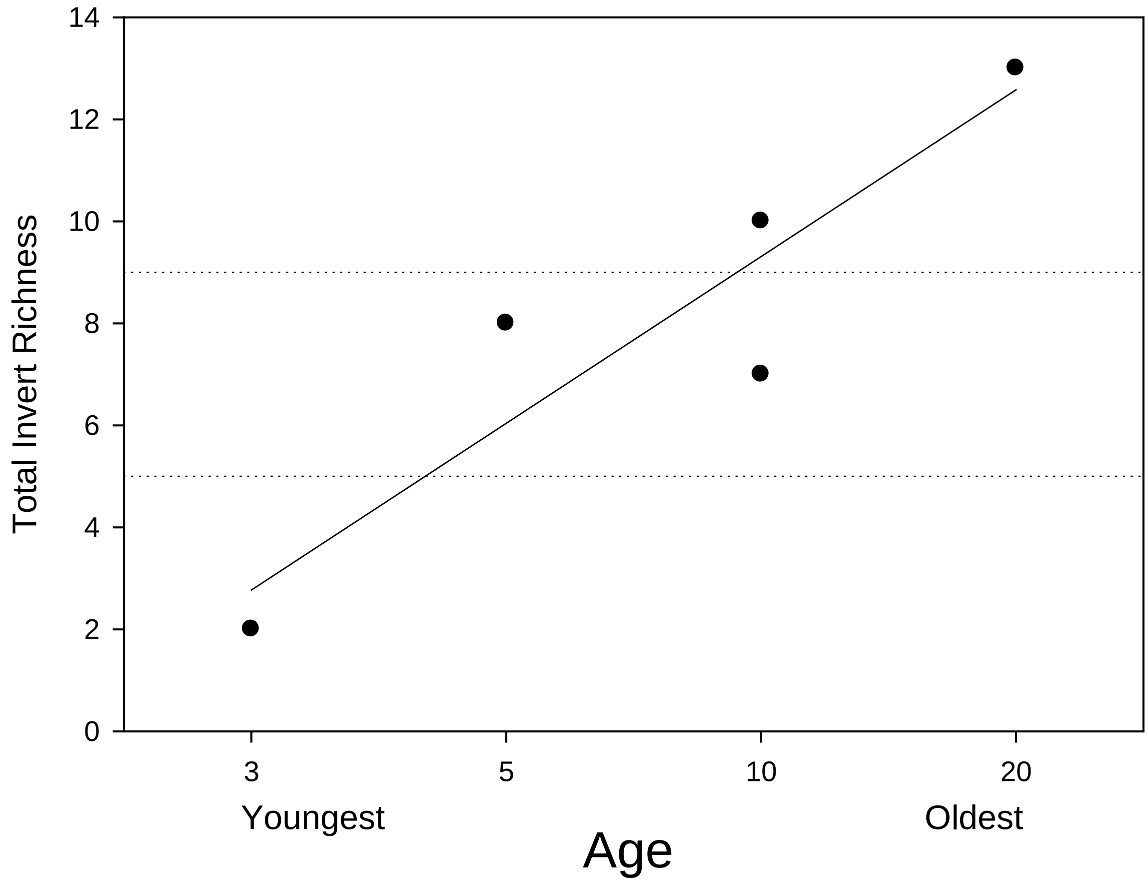

Figure 60. Total invertebrate richness for reclaimed mine perimeter channel sites in order of age since reclamation. Confidence interval for reference sites (5 – 9) is shown (dotted line).

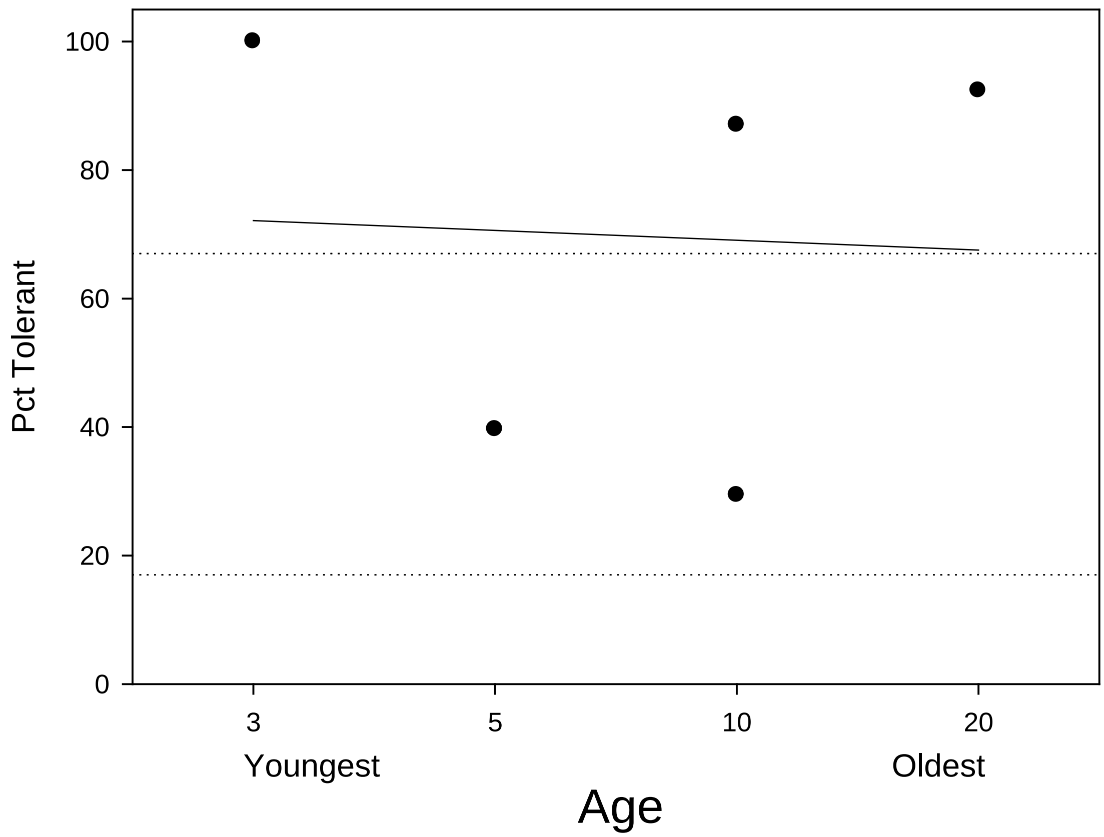

Figure 61. Percent tolerant invertebrates for reclaimed mine perimeter channel sites in order of age since reclamation. Confidence interval for reference sites (17 – 67) is shown (dotted line).

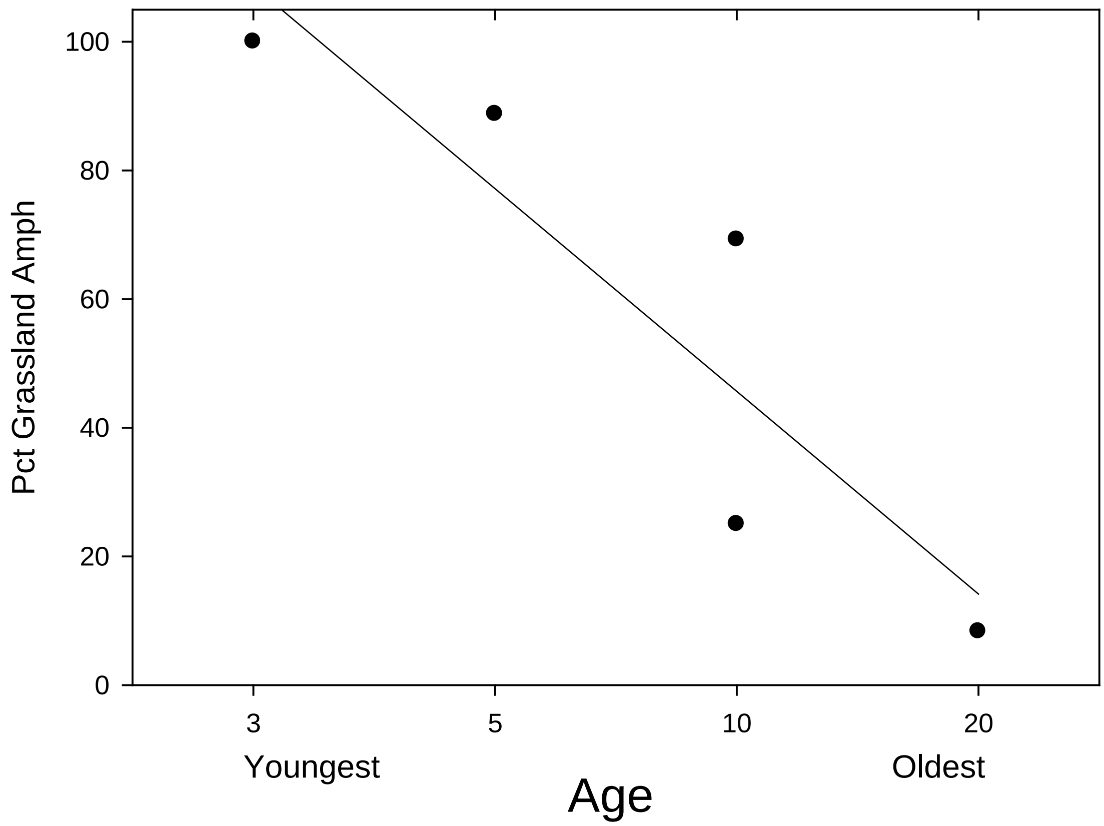

Figure 62. Percent grassland amphibians for reclaimed mine perimeter channel sites in order of age since reclamation. Confidence interval for reference sites (0) is not shown.

Appendices

Appendix A. Seasonal water chemistry measures for reference sites and reclaimed mine perimeter channels. Reference sites did not contain enough water for sampling during autumn. MDL= method detection limit. Mean and standard deviation by site type are given in the last two rows. Perimeter channel sites are listed in increasing age since reclamation.

	Season	Acidity mg/L	Alk mg/L	Cond µS/cm	Ca mg/L	Mg mg/L	SO_4 mg/L	Na mg/L	Cl mg/L
P_WO	Spring	0	181	-	241	231	1245	-	-
	Summer	0	194	3230	316	302	2131	7.43	6.9
	Autumn	0	207	3620	403	442	2120	7.66	9.2
	Winter	0	144	2905	266	276	1555	7.64	6.3
P_AR	Spring	0	139	1382	28	66	660	5.84	2.3
	Summer	0	123	1343	176	77	776	7.21	0.6
	Autumn	0	131	1428	219	95	715	8.15	2.3
	Winter	0	104	1311	185	92	652	10.11	4.3
P_ST	Spring	0	168	-	222	206	1390	20.91	4.3
	Summer	0	208	2560	246	254	1747	7.87	8.8
	Autumn	0	203	3250	404	486	2290	12.32	9.8
	Winter	0	125	1030	67	65	448	3.84	1.7
P_SU	Spring	0	114	769	40	39	282	1.83	1.4
	Summer	0	128	741	61	50	297	2.29	0.3
	Autumn	0	156	1452	161	140	695	6.27	3.3
	Winter	0	109	1937	197	194	996	-	5.7
P_BH	Spring	0	127	-	149	152	1026	40.25	43.1
	Summer	0	131	2120	155	179	1212	44.31	90.6
	Autumn	0	172	2580	263	382	1475	61.43	111.2
	Winter	0	78	1588	98	120	702	28.40	24.3
Perimeter	Spring	0 ± 0	146 ± 28	1076 ± 628	136 ± 99	139 ± 84	921 ± 451	17.21 ± 16.94	12.8 ± 18.4
	Summer	0 ± 0	157 ± 40	1999 ± 982	191 ± 96	172 ± 109	1233 ± 734	13.82 ± 17.19	21.4 ± 38.8
	Autumn	0 ± 0	174 ± 32	2466 ± 1008	290 ± 110	309 ± 179	1459 ± 752	19.17 ± 23.73	27.1 ± 47.1
	Winter	0 ± 0	112 ± 24	1754 ± 726	163± 80	150 ± 86	871 ± 430	12.50 ± 10.98	8.5 ± 9.0

Appendix A continued.

	Season	Acidity mg/L	Alk mg/L	Cond µS/cm	Ca mg/L	Mg mg/L	SO_4 mg/L	Na mg/L	Cl mg/L
R_HC	Spring	21	-	54	123	1	-	0.42	1.9
	Summer	12	6	2740	103	56	8	293.22	-
	Autumn	-	-	-	-	-	-	-	-
	Winter	-	4	462	16	12	10	44.00	102.4
R_LF	Spring	4	7	-	3	2	13	-	0.8
	Summer	-	8	52	3	2	10	0.41	0.9
	Autumn	-	-	-	-	-	-	-	-
	Winter	4	7	50	2	2	11	0.86	1.3
R_ME	Spring	6	9	67	4	3	16	1.78	1.1
	Summer	5	11	64	3	3	13	0.42	0.9
	Autumn	-	-	-	-	-	-	-	-
	Winter	7	5	89	3	3	13	4.76	1.1
R_MW	Spring	4	6	47	3	2	17	2.07	1.0
	Summer	6	8	51	2	2	11	0.54	0.9
	Autumn	-	-	-	-	-	-	-	-
	Winter	8	2	44	2	2	13	2.41	1.1
R_WO	Spring	28	-	-	15	17	121	.	0.7
	Summer	35	0	356	17	20	132	0.54	3.1
	Autumn	-	-	-	-	-	-	-	-
	Winter	32	0	278	12	15	86	2.41	1.2
Reference	Spring	13 ± 11	7 ± 4	56 ± 31	30 ± 53	5 ± 7	42 ± 50	1.42 ± 1.00	1.1 ± 0.5
	Summer	14 ± 14	7 ± 4	652 ± 1174	26 ± 44	17 ± 23	35 ± 54	59.03 ± 130.92	1.4 ± 1.2
	Autumn	-	-	-	-	-	-	-	-
	Winter	13 ± 12	4 ± 3	185 ± 182	7 ± 6	7 ± 6	26 ± 33	10.89 ± 18.56	21.4 ± 45.3

Appendix A continued.

	Season	Al mg/L	Fe mg/L	Se mg/L	Zn mg/L	Cd mg/L	Cr mg/L	Co mg/L
P_WO	Spring	0.065	0.01	MDL	MDL	MDL	MDL	0.016
	Summer	0.068	0.02	MDL	0.053	MDL	MDL	0.021
	Autumn	0.130	1.06	0.148	0.141	MDL	MDL	MDL
	Winter	0.104	0.11	0.052	0.097	0.012	0.013	0.013
P_AR	Spring	0.100	0.10	MDL	MDL	MDL	MDL	MDL
	Summer	0.085	0.15	MDL	MDL	MDL	MDL	MDL
	Autumn	0.100	0.54	MDL	MDL	MDL	MDL	MDL
	Winter	0.036	0.05	MDL	MDL	MDL	MDL	MDL
P_ST	Spring	0.088	0.04	MDL	MDL	MDL	MDL	MDL
	Summer	0.053	0.08	MDL	MDL	MDL	MDL	MDL
	Autumn	0.100	1.11	0.173	0.071	0.057	0.066	0.054
	Winter	0.061	0.07	MDL	0.018	MDL	MDL	0.013
P_SU	Spring	0.100	0.10	MDL	MDL	MDL	MDL	MDL
	Summer	0.072	0.64	MDL	MDL	MDL	MDL	0.018
	Autumn	0.100	0.69	MDL	MDL	MDL	MDL	MDL
	Winter	0.037	0.03	MDL	0.035	MDL	MDL	MDL
P_BH	Spring	0.056	0.01	MDL	MDL	MDL	MDL	0.016
	Summer	0.065	0.07	MDL	0.020	MDL	MDL	MDL
	Autumn	0.100	1.02	MDL	0.115	0.023	0.025	0.021
	Winter	0.069	0.08	MDL	0.023	MDL	MDL	0.013
Perimeter	Spring	0.082 ± 0.020	0.05 ± 0.04	-	-	-	-	0.016 ± 0.009
	Summer	0.069 ± 0.012	0.19 ± 0.25	-	0.037 ± 0.023	-	-	0.020 ± 0.011
	Autumn	0.106 ± 0.013	0.88 ± 0.25	0.161 ± 0.088	0.109 ± 0.065	0.040 ± 0.025	0.046 ± 0.029	0.038 ± 0.024
	Winter	0.061 ± 0.028	0.07 ± 0.03	0.052 ± 0.023	0.043 ± 0.037	0.012 ± 0.005	0.013 ± 0.006	0.013 ± 0.007

Appendix A continued.

	Season	Al mg/L	Fe mg/L	Se mg/L	Zn mg/L	Cd mg/L	Cr mg/L	Co mg/L
R_HC	Spring	0.100	0.10	MDL	MDL	MDL	0.016	MDL
	Summer	0.155	0.12	MDL	MDL	MDL	MDL	MDL
	Autumn	-	-	-	-	-	-	-
	Winter	MDL	0.01	MDL	MDL	MDL	MDL	MDL
R_LF	Spring	MDL	0.01	MDL	MDL	MDL	MDL	0.018
	Summer	0.051	0.10	MDL	MDL	MDL	MDL	MDL
	Autumn	-	-	-	-	-	-	-
	Winter	0.085	0.06	MDL	0.018	0.012	0.020	0.013
R_ME	Spring	0.100	0.10	MDL	0.020	MDL	MDL	MDL
	Summer	0.072	0.09	MDL	MDL	MDL	MDL	MDL
	Autumn	-	-	-	-	-	-	-
	Winter	MDL	0.01	MDL	MDL	MDL	MDL	MDL
R_MW	Spring	0.100	0.10	MDL	0.020	MDL	MDL	MDL
	Summer	0.023	0.07	0.063	0.028	MDL	0.013	MDL
	Autumn	-	-	-	-	-	-	-
	Winter	MDL	0.01	MDL	MDL	MDL	MDL	MDL
R_WO	Spring	3.000	0.01	0.053	0.458	MDL	MDL	-
	Summer	3.139	0.01	0.063	0.028	MDL	MDL	MDL
	Autumn	-	-	-	-	-	-	-
	Winter	1.308	0.10	MDL	MDL	0.012	0.015	0.016
Reference	Spring	0.825 ± 1.309	0.07 ± 0.05	0.053 ± 0.024	0.166 ± 0.201	-	0.016 ± 0.007	0.018 ± 0.008
	Summer	0.069 ± 1.371	0.08 ± 0.04	0.063 ± 0.035	0.028 ± 0.002	-	0.013 ± 0.006	-
	Autumn	-	-	-	-	-	-	-
	Winter	0.696 ± 0.569	0.04 ± 0.04	-	0.018 ± 0.008	0.012 ± 0.007	0.017 ± 0.010	0.014 ± 0.008

Appendix A continued.

	Season	Cu mg/L	Ba mg/L	Mn mg/L	Ni mg/L	NO_2 mg/L	NO_3 mg/L	NH_3 mg/L	TP mg/L
P_WO	Spring	MDL	0.02	0.02	0.05	MDL	MDL	MDL	MDL
	Summer	MDL	0.02	0.19	0.13	0.40	143.83	0.033	0.05
	Autumn	MDL	MDL	0.19	0.06	0.10	79.27	MDL	MDL
	Winter	MDL	MDL	0.42	0.14	0.07	13.53	0.009	0.68
P_AR	Spring	MDL	0.01	0.10	MDL	MDL	2.28	MDL	MDL
	Summer	MDL	0.02	0.23	MDL	0.03	1.01	0.007	0.05
	Autumn	MDL	0.02	0.16	MDL	MDL	0.24	-	MDL
	Winter	MDL	0.02	0.05	MDL	MDL	2.25	MDL	0.07
P_ST	Spring	MDL	0.02	0.04	0.03	MDL	0.74	MDL	MDL
	Summer	MDL	0.02	0.13	MDL	0.03	-	0.039	0.05
	Autumn	0.061	0.07	0.10	0.06	MDL	0.70	0.087	0.07
	Winter	MDL	0.02	0.06	0.02	MDL	0.17	MDL	MDL
P_SU	Spring	MDL	0.02	0.10	MDL	MDL	0.06	0.002	0.03
	Summer	MDL	0.03	3.13	0.04	0.03	0.02	0.009	0.05
	Autumn	MDL	0.04	0.10	MDL	MDL	MDL	MDL	MDL
	Winter	MDL	0.02	0.02	MDL	MDL	-	MDL	0.06
P_BH	Spring	MDL	MDL	0.24	MDL	MDL	MDL	MDL	MDL
	Summer	MDL	0.01	0.10	MDL	0.03	0.02	0.003	0.05
	Autumn	0.027	0.02	0.10	MDL	MDL	0.03	MDL	0.05
	Winter	MDL	MDL	0.07	0.02	MDL	0.93	MDL	0.08
Perimeter	Spring	-	0.02 ± 0.01	0.10 ± 0.08	0.04 ± 0.02	-	1.03 ± 0.98	0.002 ± 0.001	0.03 ± 0.01
	Summer	-	0.02 ± 0.01	0.75 ± 1.33	0.09 ± 0.06	0.10 ± 0.17	36.22 ± 64.21	0.018 ± 0.017	0.05 ± 0.00
	Autumn	0.044 ± 0.027	0.04 ± 0.03	0.13 ± 0.04	0.06 ± 0.03	0.10 ± 0.04	20.06 ± 35.34	0.087 ± 0.039	0.06 ± 0.03
	Winter	-	0.02 ± 0.01	0.12 ± 0.17	0.19 ± 0.06	0.07 ± 0.03	4.22 ± 5.75	0.009 ± 0.004	0.22 ± 0.28

Appendix A continued.

	Season	Cu mg/L	Ba mg/L	Mn mg/L	Ni mg/L	NO_2 mg/L	NO_3 mg/L	NH_3 mg/L	TP mg/L
R_HC	Spring	MDL	0.01	0.10	MDL	MDL	0.11	0.006	0.03
	Summer	MDL	0.89	0.36	0.03	46.23	1.85	0.056	0.05
	Autumn	-	-	-	-	-	-	-	-
	Winter	MDL	0.10	0.02	MDL	MDL	0.97	MDL	MDL
R_LF	Spring	MDL	0.03	0.02	MDL	MDL	0.24	MDL	0.06
	Summer	MDL	0.03	0.03	MDL	0.06	1.61	0.012	0.05
	Autumn	-	-	-	-	-	-	-	-
	Winter	MDL	0.06	0.06	0.02	MDL	0.57	0.029	0.10
R_ME	Spring	MDL	0.03	0.10	MDL	MDL	1.06	0.009	0.08
	Summer	MDL	0.04	0.04	MDL	0.03	1.79	0.019	0.05
	Autumn	-	-	-	-	-	-	-	-
	Winter	MDL	0.02	0.02	MDL	MDL	0.69	MDL	0.06
R_MW	Spring	MDL	0.03	0.10	MDL	MDL	MDL	0.010	MDL
	Summer	MDL	0.03	0.02	MDL	0.03	0.29	0.026	0.06
	Autumn	-	-	-	-	-	-	-	-
	Winter	MDL	0.01	0.02	MDL	MDL	0.12	MDL	0.08
R_WO	Spring	MDL	0.07	1.87	0.19	MDL	0.23	MDL	0.05
	Summer	MDL	0.04	1.90	MDL	0.03	1.52	0.026	0.05
	Autumn	-	-	-	-	-	-	-	-
	Winter	MDL	0.03	1.10	MDL	MDL	0.43	0.003	MDL
Reference	Spring	-	0.03 ± 0.02	0.44 ± 0.80	0.03 ± 0.08	-	0.41 ± 0.42	0.008 ± 0.005	0.05 ± 0.03
	Summer	-	0.20 ± 0.38	0.47 ± 0.81	0.02 ± 0.01	9.28 ± 20.66	1.41 ± 0.64	0.028 ± 0.017	0.05 ± 0.01
	Autumn	-	-	-	-	-	-	-	-
	Winter	-	0.04 ± 0.03	0.24 ± 0.48	0.02 ± 0.01	-	0.56 ± 0.32	0.016 ± 0.012	0.08 ± 0.04

Appendix B. Seasonal temperature data for reference sites and perimeter channels for periods when streams contained water. Mean and standard deviation by site type are given in the last two rows. Perimeter channel sites are listed in order of increasing age since reclamation.

Site Code	Season	Max Daily Temp (°C)	Min Daily Temp (°C)	Mean Daily Temp (°C)	CV for Mean Daily Temp
P_WO	Spring	-	-	-	-
	Summer	-	-	-	-
	Autumn	-	-	-	-
	Winter	-	-	-	-
P_AR	Spring	20.5	5.3	13.9	23.0
	Summer	22.1	15.4	18.3	5.3
	Autumn	16.6	3.9	9.0	40.9
	Winter	13.4	0.4	5.0	53.0
P_ST	Spring	26.1	6.3	16.7	23.2
	Summer	28.2	13.8	20.2	7.8
	Autumn	18.2	1.4	8.5	52.2
	Winter	19.2	0.1	5.5	72.6
P_SU	Spring	40.6	5.3	15.6	26.7
	Summer	29.1	15.8	22.6	6.4
	Autumn	22.1	0.0	9.6	55.4
	Winter	16.4	0.0	5.1	59.2
P_BH	Spring	34.6	2.0	16.1	27.6
	Summer	30.3	13.4	21.0	8.2
	Autumn	19.9	0.0	8.1	66.9
	Winter	22.3	0.0	3.7	112.7
Perimeter	Spring	30.4 ± 15.6	4.7 ± 2.7	15.6 ± 7.0	-
	Summer	27.4 ± 12.7	14.6 ± 6.6	20.5 ± 9.3	-
	Autumn	19.2 ± 8.8	1.3 ± 1.7	8.8 ± 4.0	-
	Winter	17.8 ± 8.6	0.1 ± 0.2	4.8 ± 2.3	-

Appendix B continued.

Site Code	Season	Max Daily Temp (°C)	Min Daily Temp (°C)	Mean Daily Temp (°C)	CV for Mean Daily Temp
R_HC	Spring	33.2	10.0	17.4	15.6
	Summer	38.9	11.7	20.1	6.5
	Autumn	18.2	4.2	10.0	31.3
	Winter	13.9	0.0	8.1	34.3
R_LF	Spring	31.4	2.7	12.9	33.0
	Summer	29.3	10.5	19.6	9.0
	Autumn	23.9	0.0	7.8	66.0
	Winter	10.7	0.0	4.1	69.0
R_ME	Spring	22.5	6.2	11.8	23.9
	Summer	24.3	11.0	18.3	8.3
	Autumn	19.5	0.0	8.3	51.0
	Winter	10.6	0.0	5.3	42.4
R_MW	Spring	34.1	1.7	13.2	32.1
	Summer	16.1	0.0	5.2	63.9
	Autumn	6.6	3.8	5.0	15.5
	Winter	17.4	0.0	5.2	62.2
R_WO	Spring	28.2	13.4	15.9	6.4
	Summer	32.2	12.3	19.0	10.4
	Autumn	24.9	0.0	8.9	55.4
	Winter	15.8	0.0	4.0	71.2
Reference	Spring	29.9 ± 4.7	6.8 ± 4.9	14.3 ± 2.3	-
	Summer	28.2 ± 8.6	9.1 ± 5.1	16.4 ± 6.3	-
	Autumn	18.6 ± 7.3	1.6 ± 2.2	8.0 ± 1.9	-
	Winter	13.2 ± 6.5	0.0 ± 10.0	5.7 ± 2.9	-

Appendix C. Adult amphibian abundance totals observed on reclaimed mine perimeter channels and reference streams for four sample periods. Totals by site type are given in the last two columns. Totals by site are given in the last two rows. Perimeter channel sites are listed in increasing age since reclamation.

Species	P_WO	P_AR	P_ST	P_SU	P_BH	R_HC	R_LF	R_ME	R_MW	R_WO	Perimeter	Reference
Desmognathus fuscus	0	0	0	0	0	3	22	23	10	0	0	58
Desmognathus monticola	0	0	0	0	0	5	20	46	4	0	0	75
Desmognathus unknown	0	0	0	0	0	0	1	0	1	0	0	2
Eurycea bislineata	0	0	0	0	0	0	1	0	0	1	0	2
Gyrinophilus porphyriticus	0	0	0	0	0	0	1	0	0	0	0	1
Notophthalmus v. viridescens	5	1	0	1	0	0	0	0	0	0	7	0
Pseudacris c. crucifer	0	0	0	0	0	0	0	0	0	1	0	1
Rana catesbeiana	0	0	1	0	0	0	0	0	0	0	1	0
Rana clamitans	0	1	1	0	0	0	0	0	0	0	2	0
Rana palustris	1	0	1	0	0	0	0	0	0	0	2	0
Rana sp.	0	15	0	0	0	0	0	0	0	0	15	0
Total Individuals	6	17	3	1	0	8	45	69	15	2	27	139
Total Species	2	3	3	1	0	2	5	2	3	2	5	6

Appendix D. Larval amphibian abundance survey totals observed on reclaimed mine perimeter channels and reference streams for four sample periods. Totals by site type are given in the last two columns. Totals by site are given in the last two rows. Perimeter channel sites are listed in increasing age since reclamation.

Species	P_WO	P_AR	P_ST	P_SU	P_BH	R_HC	R_LF	R_ME	R_MW	R_WO	Perimeter	Reference
Ambystoma sp.	0	1	0	0	0	0	0	0	0	0	1	0
Bufo americana	0	0	0	3	0	0	0	0	0	0	3	0
Desmognathus fuscus	0	0	0	0	0	3	5	12	2	1	0	23
Eurycea cirrigera	0	0	0	0	0	0	3	1	12	0	0	16
Hyla chrysoscelis	0	1	0	1	11	0	0	0	0	0	13	0
Notophthalmus v. viridescens	0	15	0	5	0	0	0	0	0	0	20	0
Pseudacris c. crucifer	0	7	9	3	0	0	0	0	0	0	19	0
Rana clamitans	0	39	0	0	1	0	0	0	0	0	40	0
Total Individuals	0	63	9	12	12	3	8	13	14	1	96	39
Total Species	0	5	1	4	2	1	2	2	2	1	6	2

Appendix E. Combined larval and adult amphibian abundance survey totals observed on reclaimed mine perimeter channels and reference sites for four sample periods. Totals by site type are given in the last two columns. Totals by site are given in the last two rows. Perimeter channel sites are listed in order of increasing age since reclamation.

Species	P_WO	P_AR	P_ST	P_SU	P_BH	R_HC	R_LF	R_ME	R_MW	R_WO	Perimeter	Reference
Ambystoma sp.	0	1	0	0	0	0	0	0	0	0	1	0
Bufo americana	0	0	0	3	0	0	0	0	0	0	3	0
Desmognathus fuscus	0	0	0	0	0	6	27	35	12	1	0	81
Desmognathus monticola	0	0	0	0	0	5	20	46	4	0	0	75
Desmognathus unknown	0	0	0	0	0	0	1	0	1	0	0	2
Eurycea bislineata	0	0	0	0	0	0	1	0	0	1	0	2
Eurycea cirrigera	0	0	0	0	0	0	3	1	12	0	0	16
Gyrinophilus porphyriticus	0	0	0	0	0	0	1	0	0	0	0	1
Hyla chrysoscelis	0	1	0	1	11	0	0	0	0	0	13	0
Notophthalmus v. viridescens	5	16	0	6	0	0	0	0	0	0	27	0
Pseudacris c. crucifer	0	7	9	3	0	0	0	0	0	1	19	1
Rana catesbeiana	0	0	1	0	0	0	0	0	0	0	1	0
Rana clamitans	0	40	1	0	1	0	0	0	0	0	42	0
Rana palustris	1	0	1	0	0	0	0	0	0	0	2	0
Rana sp.	0	15	0	0	0	0	0	0	0	0	15	0
Total Individuals	6	80	12	13	12	11	53	82	29	3	123	178
Total Species	2	6	4	4	2	2	6	3	4	3	9	7

Appendix F. Macroinvertebrate abundance data given by Order (when known) for reclaimed mine perimeter channels and reference sites. Mean and standard deviation by site type are given in the last two columns. Perimeter channel sites are listed in order of increasing age since reclamation.

Order	P_WO	P_AR	P_ST	P_SU	P_BH	R_HC	R_LF	R_ME	R_MW	R_WO	Perimeter	Reference
Cladocera	0	0	210	0	0	0	0	1	16	0	42 ± 94	3 ± 7
Clams	0	0	8	0	16	0	0	0	0	0	5 ± 7	0 ± 0
Coleoptera	0	8	15	0	3	0	0	0	0	0	5 ± 7	0 ± 0
Collembola	0	0	0	0	24	0	0	8	56	0	5 ± 11	13 ± 24
Cyclopoida	0	0	88	96	0	8	0	0	0	0	37 ± 50	2 ± 4
Diptera	1329	33	56	1196	705	20	16	68	184	41	664 ± 611	66 ± 69
Ephemeroptera	0	16	16	2	0	0	88	15	128	0	7 ± 8	46 ± 58
Hemiptera	0	0	2	0	0	0	0	0	16	0	0 ± 1	3 ± 7
Odonata	0	10	0	6	3	0	0	1	0	0	4 ± 4	0 ± 0
Oligochaeta	0	1	0	8	56	8	0	0	8	8	13 ± 24	5 ± 4
Plecoptera	0	0	0	1	0	3	41	208	135	11	0 ± 0	80 ± 89
Snails	0	18	8	75	16	8	0	0	0	0	23 ± 30	2 ± 4
Trichoptera	0	0	0	0	0	0	0	1	1	0	0 ± 0	0 ± 1

Appendix G. Macroinvertebrate abundance data given as a percent by Order (when known) for reclaimed mine perimeter channels and reference streams. Mean and standard deviation by site type are given in the last two columns. Perimeter channel sites are listed in order of increasing age since reclamation.

Order	P_WO	P_AR	P_ST	P_SU	P_BH	R_HC	R_LF	R_ME	R_MW	R_WO	Perimeter	Reference
Cladocera	0	0	52	0	0	0	0	0	3	0	10 ± 23	1 ± 1
Clams	0	0	2	0	2	0	0	0	0	0	1 ± 1	0 ± 0
Coleoptera	0	9	4	0	0	0	0	0	0	0	3 ± 4	0 ± 0
Collembola	0	0	0	0	3	0	0	3	10	0	1 ± 1	3 ± 4
Cyclopoida	0	0	22	7	0	17	0	0	0	0	6 ± 9	3 ± 8
Diptera	100	38	14	86	86	43	11	23	34	68	65 ± 37	36 ± 22
Ephemeroptera	0	19	4	0	0	0	61	5	24	0	5 ± 8	18 ± 26
Hemiptera	0	0	0	0	0	0	0	0	3	0	0 ± 0	1 ± 1
Odonata	0	12	0	0	0	0	0	0	0	0	2 ± 5	0 ± 0
Oligochaeta	0	1	0	1	7	17	0	0	1	13	2 ± 3	6 ± 8
Plecoptera	0	0	0	0	0	6	28	69	25	18	0 ± 0	29 ± 24
Snails	0	21	2	5	2	17	0	0	0	0	6 ± 9	3 ± 8
Trichoptera	0	0	0	0	0	0	0	0	0	0	0 ± 0	0 ± 0

Appendix H. Macroinvertebrate abundance data given by Genus (when known) for reclaimed mine perimeter channels and reference streams. Mean and standard deviation by site type are given in the last two columns. Perimeter channel sites are listed in order of increasing age since reclamation.

Class/Order	Genera	P_WO	P_AR	P_ST	P_SU	P_BH	R_HC	R_LF	R_ME	R_MW	R_WO	Perimeter	Reference
Oligochaeta	-	0	1	0	8	56	8	0	0	8	8	13 ± 24	5 ± 4
Bivalvia (clam)	-	0	0	8	0	16	0	0	0	0	0	5 ± 7	0 ± 0
Gastropoda (snail)	-	0	18	8	75	16	8	0	0	0	0	23 ± 30	2 ± 4
Ephemeroptera	Baetis	0	3	0	0	0	0	11	0	0	0	1 ± 1	2 ± 5
Ephemeroptera	Centroptilum	0	13	0	0	0	0	0	0	0	0	3 ± 6	0 ± 0
Ephemeroptera	Acerpenna	0	1	0	0	0	0	0	0	0	0	0 ± 0	0 ± 0
Ephemeroptera	Ephemerellidae(UNK)	0	2	16	0	0	0	0	7	0	0	4 ± 7	1 ± 3
Ephemeroptera	Ephemerella	0	0	0	0	0	0	2	0	35	0	0 ± 0	7 ± 15
Ephemeroptera	Ephemera	0	0	0	0	0	0	0	0	5	0	0 ± 0	1 ± 2
Ephemeroptera	Ameletus	0	0	0	2	0	0	67	0	102	0	0 ± 1	34 ± 48
Ephemeroptera	Mayfly(UNK)	0	0	0	0	0	0	8	8	0	0	0 ± 0	3 ± 4
Trichoptera	Hydropsyche	0	0	0	0	0	0	0	0	1	0	0 ± 0	0 ± 0
Trichoptera	Caddisfly(UNK)	0	0	0	0	0	0	0	1	0	0	0 ± 0	0 ± 0
Plecoptera	Capnia	0	0	0	0	0	0	1	0	0	0	0 ± 0	0 ± 0
Plecoptera	Leuctridae(UNK)	0	0	0	0	0	3	0	49	74	11	0 ± 0	27 ± 33
Plecoptera	Leuctra	0	0	0	0	0	0	12	0	0	0	0 ± 0	2 ± 5
Plecoptera	Capniidae/Leuctridae(UNK)	0	0	0	0	0	0	0	0	16	0	0 ± 0	3 ± 7
Plecoptera	Perlodidae(UNK)	0	0	0	0	0	0	0	0	16	0	0 ± 0	3 ± 7
Plecoptera	Isoperla	0	0	0	0	0	0	12	0	0	0	0 ± 0	2 ± 5
Plecoptera	Yugus	0	0	0	1	0	0	0	0	0	0	0 ± 0	0 ± 0
Plecoptera	Peltoperla	0	0	0	0	0	0	16	150	0	0	0 ± 0	33 ± 66
Plecoptera	Nemouridae(UNK)	0	0	0	0	0	0	0	9	3	0	0 ± 0	2 ± 4

Appendix H continued.

Class/Order	Genera	P_WO	P_AR	P_ST	P_SU	P_BH	R_HC	R_LF	R_ME	R_MW	R_WO	Perimeter	Reference
Odonata	Gomphidae(UNK)	0	8	0	0	0	0	0	0	0	0	2 ± 4	0 ± 0
Odonata	Libellulidae	0	2	0	3	0	0	0	1	0	0	1 ± 1	0 ± 0
Odonata	Coenagrionidae	0	0	0	3	3	0	0	0	0	0	1 ± 2	0 ± 0
Coleoptera	Dytiscidae(UNK)	0	0	15	0	1	0	0	0	0	0	3 ± 7	0 ± 0
Coleoptera	Agabus	0	0	0	0	2	0	0	0	0	0	0 ± 1	0 ± 0
Coleoptera	Peltodytes	0	8	0	0	0	0	0	0	0	0	2 ± 4	0 ± 0
Diptera	Chironomidae	1246	0	55	1112	0	0	0	0	0	24	483 ± 638	5 ± 11
Diptera	Tipulidae(UNK)	0	0	0	0	1	1	0	0	0	0	0 ± 0	0 ± 0
Diptera	Tabanus	0	0	0	0	0	0	8	0	0	0	0 ± 0	2 ± 4
Diptera	Chrysops	0	0	0	0	1	0	0	0	0	0	0 ± 0	0 ± 0
Diptera	Simulium	0	0	0	2	49	0	0	0	0	0	10 ± 22	0 ± 0
Diptera	Ceratopogonidae(UNK)	83	8	1	0	0	0	0	0	0	0	18 ± 36	0 ± 0
Diptera	Bezzia	0	0	0	0	1	0	0	0	0	0	0 ± 0	0 ± 0
Diptera	Stratiomyidae	0	0	0	4	0	0	0	0	0	0	1 ± 2	0 ± 0
Diptera	Tanyderidae	0	0	0	0	2	0	0	0	0	0	0 ± 1	0 ± 0
Diptera	Diptera(UNK)	0	16	0	8	2	1	8	8	0	0	5 ± 7	3 ± 4
Diptera	Non-Tanypodinae	0	9	0	35	593	18	0	51	176	17	127 ± 261	52 ± 72
Diptera	Tanypodinae	0	0	0	35	56	0	0	0	0	0	18 ± 26	0 ± 0
Collembola	Sminthuridae(UNK)	0	0	0	0	0	0	0	8	0	0	0 ± 0	2 ± 4
Collembola	Sminthurides	0	0	0	0	0	0	0	0	56	0	0 ± 0	11 ± 25
Collembola	Agrenia bidenticulata	0	0	0	0	24	0	0	0	0	0	5 ± 11	0 ± 0
Cyclopoida	Cladocera	0	0	210	0	0	0	0	0	16	0	42 ± 94	3 ± 7
Hemiptera	Hemiptera(UNK)	0	0	2	0	0	0	0	0	16	0	0 ± 1	3 ± 7
Hemiptera	Mesouelia	0	0	0	0	0	0	0	0	0	0	0 ± 0	0 ± 0
Decapoda	Crayfish(UNK)	0	0	0	0	0	0	0	1	0	0	0 ± 0	0 ± 0
Coleoptera	Hydrocanthus	0	0	0	0	0	0	0	0	0	0	3 ± 0	0 ± 0
Calanoida	Copepod(UNK)	0	0	88	96	0	16	0	0	0	0	37 ± 50	3 ± 7